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CLINICAL AND EXPERIMENTAL STUDIES ON SODIUM BENZOATE

THE VALUE OF THE SODIUM BENZOATE TEST OF RENAL FUNCTION, AND THE EFFECT OF INJURY OF THE LIVER ON HIPPUIC ACID SYNTHESIS¹

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(Received for publication February 26, 1925)

INTRODUCTION

These studies were undertaken in order to determine the clinical value of the sodium benzoate test of renal function as proposed by Kingsbury and Swanson (27)

Hippuric acid has claimed the attention of physiologic chemists and pathologists for many years (40). The basis of this interest lies in the generally accepted fact of its formation in the body by the conjugation of benzoic acid, or its salts, with glycoll, resulting in the detoxication of the former and the loss of the latter to the body. The question as to where this conjugation takes place and whence comes the large amount of requisite glycoll still claims the attention of the chemist. The interest of the pathologist is due to the celebrated experiments of Bunge and Schmiedeberg (8), who, in 1877, perfused the kidneys of surviving dogs with benzoic acid and glycoll, and later found hippuric acid in the transfused blood, from which they concluded that, at least in the dog, the kidney is the site of the synthesis. For many years their results stood unchallenged. If correct, this function of the kidney is singular since other substances synthesized by the body appear in the various organs, but are only excreted by the kidney. The possible aberration of this synthesizing

¹This work was done under the direction of Dr L. G. Rowntree, of the Division of Medicine, Mayo Clinic.

ability in the case of diseased kidneys is the basis of numerous attempts to employ it as a test of renal function

The employment of sodium benzoate in determining renal function is not new. As early as 1879 Jaarsvald and Stokvis (22) studied a number of cases with various types of renal complaints. They concluded that in man the synthesis of hippuric acid is diminished or entirely absent in affections of the kidney, more especially in parenchymatous nephritis. In 1880 Weyl and Anrep (50), and in 1883, Kronecker (28) carried on rather similar work with somewhat contradictory results. Kingsbury has explained these by the fact that with their method some of the hippuric acid was hydrolyzed to benzoic acid by alkali. More recently Violle (49) has reported that in nephretic patients, after giving 0.5 gm doses of sodium benzoate, the excretion of hippuric acid is much less than in normal persons.

Kingsbury and Swanson (26) perfected and simplified the method of hippuric acid determination, and also administered sodium benzoate in 2.4 gm doses to many normal and abnormal persons. Their results, working with approximately normal university students, are of particular value in indicating what the average rate of synthesis should be in normal persons.

Morgulis, Pratt, and Jahr (39) carried on a somewhat parallel investigation giving particular attention to the rôle of the free benzoic acid excreted. They raised the question as to whether there was sufficient of this substance to vitiate the use of the test clinically. This was at variance with Kingsbury (25), who was able to find only traces of free benzoic acid when accidental hydrolysis had been prevented.

Snapper and his co-workers (45, 46, 47, 48) have recently reported interesting and significant work in which they not only amplified the work of Bunge and Schmiedeberg and also Kingsbury and Bell (24), but have made a number of clinical observations as well.

The earlier portion of this study, especially that dealing with pregnant and postpartum women, was carried on in one of the smaller outlying hospitals of Minneapolis, and the data, while essentially correct, are less complete than those of more recent study. Most of the work was conducted at the Mayo Clinic where it was possible to make parallel studies of the various functions.

MECHANISM OF THE SYNTHESIS

The following facts concerning the mechanism of hippuric acid synthesis seem well supported. Hippuric acid (18) is one of the nitrogenous end-products of protein metabolism and is a combination product arising by the union of glycocoll and benzoic acid.

The benzoic acid component arises from one of two sources, etc (18) either from aromatic products of vegetable food, as cinamic acid, quinic acid, and so forth, which in metabolism are catabolized to benzoic acid, or from phenylalanin, which undergoes complete dissociation in normal metabolism. The phenylpropionic acid which is produced from this in intestinal putrefaction is oxidized freely into benzoic acid when resorbed (3).

The origin of the glycocoll component is, however, far more complicated. In carnivorous animals, Brugsch and Hirsch (6) (7) found that the degree of hippuric acid formation after the introduction of benzoic acid was lower than in the herbivorous animal, that the amount of free benzoic acid was greater than that paired with glycocoll, and that in the dog detoxication of benzoic acid occurs only in relatively small part by the formation of hippuric acid and to a considerable extent by the formation of reducing substances (glycuronates).

Schmiedeberg (44) suggested that the synthesis took place under the influence of a ferment "histozyme."

In human beings Lewinski (31) found that after the administration of benzoic acid the only mode of hippuric acid formation was by hydrolytic protein cleavage. From his work he concluded that a person on a diet rich in proteins can transform more benzoic acid to hippuric acid than otherwise, and that the appearance of reducing substances in the urine is an expression of the impoverishment of the organism in glycocoll.

Magnus-Levy (36), Wiechowski (51), Ringer (41), Abderhalden (1), and Delprat and Whipple (14), found that when benzoic acid was given to herbivora in quantity, the animal excreted one-third or more of the total nitrogen as hippuric acid. Ringer suggested that the large quantities of glycocoll might originate from the "extra-destroyed" protein, and not from protein that would have been metabolized had no benzoate been given.

Epstein and Bookman (16), found no "reserve" glycocoll in the animal body. They considered the production of glycocoll dependent upon the amount of benzoic acid present in the circulation, and that the processes involved might represent either destruction of higher amino-acids or construction from simpler bodies.

The results of Lewis (33, 34), and of McCollum and Hoagland (35) were essentially consistent. Lewis concluded that the decreased excretion of uric acid must be regarded as the result of decreased uric acid formation due to the lowering of the level of nuclear metabolism, and as an example of specific influence on nuclear metabolism rather than on general endogenous metabolism. Sodium hippurate failed to produce changes in either creatinin or uric acid elimination, and hence he concluded that the effect was due to the action of free benzoic acid and not to conjugated, non toxic hippuric acid.

Whipple (14) proposed the hypothesis that the increase in urinary nitrogen is due to the lack of available glycocholate, and that under these conditions a demand for the drug being very acute, the body protein is broken down to supply the "emergency requirement" Parker and Lusk considered the possibility of a "glycocholate reserve" which can be washed out by successive doses of benzoate

Cohn (9) (10) noted that feeding proteins and their glycocholate yielding decomposition products, counteracted the toxic effect of benzoic acid in rabbits In contrast with Wiener (52), however, he failed to note detoxification of the benzoic acid when leucin was given Friedmann and Tachau (17), and Hoffmann (21) were unable to demonstrate that amino, hydroxy, or fatty acids were precursors of glycocholate

Among the most recent investigators of the origin of glycocholate are Griffith and Lewis (19) (20) In their work with rabbits they noted that the rate of excretion of hippuric acid was notably increased by the administration of glycocholate with the sodium benzoate This, they concluded, probably represented an increased rate of synthesis due to the presence in the organism of large amounts of preformed glycocholate Their later work shows that the synthesis in rabbits is enhanced by feeding protein rich in glycocholate with the sodium benzoate, although it is not enhanced by feeding other proteins, unless glycocholate is also fed From these facts they consider it improbable that the ordinary reactions of protein metabolism produce readily available precursors of glycocholate

Csonka (11) (12) does not doubt the synthetic production of glycocholate in the normal animal Experimenting with swine he found that under the influence of benzoic acid certain reducing substances (glycuronates) were eliminated in the urine The quantity of these was largest when casein was given, and smallest when gelatin was given, pointing to the inverse relation to the preformed glycocholate content of the protein ingested In his opinion, there is an increased nitrogen elimination when benzoic acid is administered, which supports the claim that the metabolism rises after its ingestion

As to the amount of sodium benzoate which a human being is able to convert, Lewinski (31) found that a man could completely convert 20 gm of benzoic acid ingested within twelve hours without ill effect, and on a diet rich in glycocholate 40 gm of benzoic acid, of which only 10 per cent was excreted in the free state Lewis (33) gave men doses of from 6 to 10 gm of sodium benzoate of which 85 or 90 per cent was eliminated in five or six hours as hippuric acid Dakin (13) found that sodium benzoate taken by human beings in 5 to 10 gm doses for two or three days is completely converted into hippuric acid and is eliminated as such in the urine, no free benzoic acid being excreted under these conditions

Particular points to be gleaned from the foregoing are (1) normal human beings may completely convert at least 25 gm of sodium benzoate to hippuric acid without ill effect and with the appearance of only traces of free benzoic acid in the urine When larger amounts of benzoic acid or its salts are given, the amount converted is greater if there is extra available glycocholate in the body, and (2) the results of many investigations point to two available sources for the origin of

glycocoll in hippuric acid formation synthesis and preformed glycocoll, the latter includes glycocoll liberated by the catabolism of body protein and that present in food protein

SITE OF THE SYNTHESIS

Important as the foregoing considerations are to a proper understanding of benzoic acid detoxication, it is even more essential to decide whether the synthesis takes place in the kidney only, or also in some other organ, such as the liver

As early as 1857 Kühne and Hallwachs (29) concluded that the formation of hippuric acid from introduced benzoic acid did not occur in the intestines, nor in the circulating blood, but in the hepatic vessels in the presence of the constituents of bile (glycocholic acid). This was supported by Rosenberg (43), and by Lewis (32) in his more recent experiment in which he inserted a cannula in the common bile duct of a rabbit and demonstrated that in this animal synthesis of hippuric acid occurred after exclusion of bile from the intestine

The perfusion experiments of Bunge and Schmiedeberg (8) have already been referred to. Their results were confirmed by Kochs (27), who noted that synthesis took place even in the comminuted kidney of the dog, ox, and calf, also by Bashford and Cramer (2) who determined hippuric acid crystals in renal tissue and blood which had been triturated until the microscope showed no intact cells, and concluded that the formation of the hippuric acid was not dependent on intact and living cells

In 1911, Friedmann and Tachau (17) experimenting with rabbit livers concluded that, at least in herbivora, the liver partook in the synthesis. Except for this, the results of Bunge and Schmiedeberg (8) were practically unchallenged until Kingsbury and Bell (24) found hippuric acid in the tissues of nephrectomized dogs after injections of glycocoll and sodium benzoate, and concluded that in the dog the kidney was not the only organ which synthesized hippuric acid

Lackner, Levinson and Morse (30) believed that in the dog the excretion of hippuric acid varied with the condition of the liver, the condition of the kidney remaining constant, and that if the liver partook in this synthesis to any great extent, pathologic involvement of the liver should be recognizable by this means. Some experimental and clinical evidence was found in support of this claim

Delprat and Whipple (14), working with dogs with necrosis of the liver induced by chloroform, concluded that in this condition the synthesis was not prevented but was distinctly delayed, also that the liver normally partakes in this action but that other cell protoplasm may partake in the reaction

Morgulis (39) and his co-workers believed that extensive injury to the liver was responsible for the failure of certain preliminary steps in the process, namely, the formation of glycocoll, rather than that it indicated an actual participation of the liver in the synthesis

Snapper, Grünbaum and Neuberg (48) believed that Kingsbury and Bell (24) based their conclusions on too little evidence. Accordingly they repeated the

work of Bunge and Schmiedeberg (8), and also that of Kingsbury and Bell (24). They were thus able to conclude, in confirmation of the earlier work of Bunge and Schmiedeberg, and in opposition to that of Kingsbury and Bell that not only is hippuric acid synthesized locally in the kidney of a dog, but that in this animal this synthesis occurs only in the kidney.

Their further work with swine and sheep (48) is of interest in this connection. It was also their privilege to perfuse two human kidneys (48) with fair remaining function, immediately after removal, one being from a patient with renal tuberculosis and the other from a patient with tumor of the kidney. In each instance they were able to demonstrate definite amounts of hippuric acid in the perfusion blood to which sodium benzoate and glycocholate had been added. From their extensive work they concluded (1) that in the perfusion of the kidneys of dogs, swine, and sheep with benzoic acid and glycocholate, hippuric acid is synthesized, and (2) that this also occurs in the isolated human kidney.

Brakefield and Schmidt (5) found that in two rabbits whose bile ducts had been ligated only 40 per cent of the amount of hippuric acid synthesized by normal rabbits was excreted, they also observed that normal dogs able to conjugate 95 per cent of a given amount of benzoate, conjugated only 60 per cent after ligation of the common bile duct.

Kingsbury and Bell (23) worked with rabbits made nephritic with racemic tartaric acid. They concluded that if hippuric acid was synthesized in the kidney of the rabbit, it must be accomplished either by the degeneration cells of the convoluted tubules, or by the cells of the glomeruli. The latter possibility is supported by our own work, that is, in the cases of nephrosis in which the glomeruli were but little impaired, the elimination of hippuric acid was but little reduced, it was greatly reduced in cases of nephritis or of other destructive renal lesions.

Morgulis (39) regards the kidney as the essential organ of hippuric acid synthesis, but finds the amount of free benzoic acid to be increased in cases of nephritic disease. Kingsbury (25), however, believes that sodium benzoate is completely converted and eliminated as hippuric acid in individuals whose kidneys have been extensively damaged, he also believes that the synthesis of the product must be as rapid as its excretion. This is in harmony with the work of Snapper and Grünbaum (47), who found that patients with renal insufficiency synthesized sodium benzoate to hippuric acid which, however, is dammed back in the blood stream.

Reviewing the literature which we have thus briefly cited, it appears certain that whenever the requisite glycocholate and benzoic acid are available to it, the kidney is the main site of the synthesis of hippuric acid. This is shown by the work of Bunge and Schmiedeberg (8), questioned by Friedmann and Tachau (17), and by Kingsbury and Bell (24), but confirmed by Kochs (27), Bashford and Cramer (2), Snapper, Grünbaum, and Neuberger (45-48 inclusive), as well as by our own work in various cases of nephritic lesions in which the elimination of hippuric acid seemed quite proportional to the extent of the renal damage. It appears equally true that the kidney cannot perform this function satisfactorily if the liver does not function properly. Morgulis' (39) conception of the liver as

playing an important although a secondary rôle, is supported by the work of Delprat and Whipple (14), by Lackner, Levinson, and Morse (30), by Brakefield and Schmidt (5), and also by us in our work upon human cases with damaged livers. That this role is distinctly secondary is most strikingly shown by our finding that a completely hepatectomized dog can synthesize appreciable amounts of hippuric acid, and by Bashford and Cramer (2), who many years ago demonstrated the synthesis of hippuric acid *in vitro* by macerated renal tissue.

METHODS

Administration of the sodium benzoate Many of the patients in the hospital were studied according to a routine which was designed to show the total elimination of hippuric benzoic acid for the twenty-four hours preceding the three-hour test, as well as for the twenty-one hours following it. It seemed that a more adequate idea would thus be obtained of the usual hippuric and benzoic acid output, as well as some control on the subject's reaction to the test.

Régime in the management of the test In the ordinary clinical application of the test the following procedure might be followed.

Fruits are omitted from the diet for at least twenty-four hours preceding the test. Just before starting the test the patient is asked to void and this specimen is discarded. The sodium benzoate is then dissolved in its container in 100 cc. of water and drunk, the container is rinsed with 200 cc. of water which is also drunk, an hour later another 200 cc. of water is taken.

Specimens are collected at the end of the second hour and at the end of the third hour and these two specimens are analyzed for hippuric acid by the methods of Kingsbury and Swanson (26).

Breakfast is withheld until after the third-hour specimen is voided.

The value of the test being once established, the "before" and "after" periods mentioned above, as well as the free benzoic acid determinations might be dispensed with. The essential information which the test may be expected to furnish may then be obtained from the two-hour and three hour determinations, or, if the deductions from the tables which follow are correct, from the results of the two-hour specimen alone. If this cannot be established the test is probably too cumbersome for any clinical application. The dietary restriction in fruits is probably necessary since benzoic acid occurs in fruits, particularly in cranberries.

Each of the fifty-two patients in this series received 2.4 gm. of sodium benzoate by mouth, as described, the amount originally employed by Kingsbury. It is equivalent to 2.0 gm. of benzoic acid, in percentage terms of which the results of the test are calculated. Thus, if a patient excretes 1.5 gm. of benzoic acid the portion of the original dose excreted may be considered as 75 per cent. More recently Kingsbury has advocated giving 1.8 gm. of sodium benzoate for each square meter of body surface, as determined by the height and weight formula of Du Bois (15). This would be, for the average adult, essentially the amount used in this study. Therefore, it appears that in considering the power of the kidneys

of the average adult to detoxicate benzoic acid the observation of this refinement would not greatly alter the results obtained. In none of the tables was the excreted hippuric acid expressed as such, but always for the sake of simplicity, in the terms of benzoic acid. The relations of hippuric to benzoic acid may be readily seen from their molecular weights, 179 and 122 respectively, the foregoing dosage, then, would be equivalent to 2.93 gm. of hippuric acid.

Total benzoic acid The method employed in this analysis was the same as that described by Kingsbury and Swanson (26), except that only half the amounts mentioned in the reference were employed, this modification was later adopted by Kingsbury himself. Since the application of this method required the removal of protein from the urine when present, as in some of the nephritic cases, this also was done by the method described by Kingsbury and Swanson. The specimens were preserved by the use of 2 per cent nitric acid and a small amount of 10 per cent thymol in chloroform. Frequent duplicate determinations were made throughout the work. The chloroform was washed, as described.

Free benzoic acid This was determined, as described by Morgulis (39) and his co-workers, except that it was not always possible to employ 100 cc. samples of urine. To quote the details:

"For the purpose of deproteinizing, sodium hydroxide was added until the strongly acid urine reacted just faintly acid to litmus. Then 0.5 gm. of tannic acid was added to every 100 cc. of urine. A flocculent precipitate was formed in the presence of albumin, which was filtered off after the mixture had been allowed to stand for a few minutes.

Of the deproteinized urine 100 cc. samples were immediately measured out into separatory funnels and saturated with ammonium sulphate. The mixture was then strongly acidified by the addition of 1 cc. of concentrated nitric acid and extracted with four portions of freshly washed chloroform."

PRESENTATION OF CASES

The fifty-two cases in the series may be considered in five groups: (1) normal subjects, (2) pregnant and postpartum subjects, (3) subjects with essential renal involvement, (4) subjects with only slight renal involvement, (5) those with definite hepatic involvement. Certain experiments with dogs are also considered in connection with those in the latter group.

With the exception of tables 2, 3, and 4, the tables were arranged according to the value of the two-hour benzoate excretion, rather than the third-hour value, since the former seems to be more significant in indicating renal impairment. This belief is based on the observation that there is a tendency for the third-hour value so to raise the value for the whole three hours that it may approximate the

normal (85 to 90 per cent), even though the two-hour value is notably decreased. Hence, at the end of the three hours the significance of the test might be concealed if it were based on this value alone.

Group 1 Normal subjects In interpreting results it is presumed that the standards set by Kingsbury are approximately correct. After experimenting with many normal university students, as well as a number of cardiac and nephritic patients, he concluded that 70 per cent or more of the initial dose (2.4 gm.), expressed as benzoic acid, is excreted in the first two-hour period, and a total of 90 per cent or more is excreted during the entire three-hour period of the test. Recently he has said "at least 80 per cent of the hippuric acid theoretically equivalent to the ingested dose of benzoate should be found in the three-hour specimen. Sixty-seven tests made on normal medical students in this laboratory (University of Minnesota), showed an output of 80 per cent or above, 91 per cent of these sixty-seven cases showed benzoate tests above 85 per cent." The results of the tests in thirteen cases are well within the limits just outlined (table 1). This is true for the blood urea and the phenolsulphonephthalein as well as for the sodium benzoate test. The three-hour value in two of the cases seems to be above 100 per cent, this is possible when the excretory power of the kidney is high and when the diet may not have been adequately controlled. Under such conditions the benzoate available from sources other than the dose given must be considerable, assuming the technic to have been faultless otherwise. This fact does not appear to militate against the value of the test since in cases in which the excretory power of the kidney is reduced, the excretion seems to proceed at a lower but maximal figure maintained for a longer time.

The subjects of this group, while not strictly normal, gave but little evidence of renal damage. The values found bear out those established by Kingsbury as being approximately correct.

Group 2 Pregnant and postpartum subjects The subjects of this group had had infections of various types which apparently affected the kidneys, and all of these infections seem to have been reflected in the results of the renal tests.

The rôle of infection in the etiology of the nephritides is a matter of common medical knowledge, and the effect of infection on the

ability of the kidney to excrete hippuric acid has been previously noted. The correctness of these observations seems well shown by a comparison of table 2, composed of cases with normal or nearly normal values for the benzoate test, with table 3, composed of cases with definitely abnormal values.

TABLE 1
Normal renal function

Case	Milligrams for each 100 cc.		Uric acid	Non protein nitrogen	Phenolsulphonaphthalein return	Sodium benzoate return			Diagnosis
	Urea	Creatinine				Two-hour	Third hour	Total	
	Normal		per cent	per cent	per cent	per cent	per cent	per cent	
1	32	1.7	3.7		65	70.0	20.0	90.0	Malignant hypertension
2	38	1.3		38.0	80	70.4	17.7	88.1	Exophthalmic goiter
3						72.0	12.0	84.0	Normal? Hemorrhagic icterus
4						72.0	32.0	104.0	Normal pregnancy
5	53	1.3	2.4		55	72.0	12.4	84.6	Adenomatous thyroid, diseased gallbladder
6					70	73.6	21.8	95.5	Exophthalmic goiter, pregnancy, three months
7	34	1.7	3.5		30	74.8	13.6	88.5	Adenoma of thyroid
8	34	1.6		40	65	77.9	11.9	89.0	Carcinoma of adrenal
9	36	1.4		38	80	78.4	17.9	96.3	Exophthalmic goiter, myocardial degeneration
10					58	81.4	17.3	98.7	Normal puerperium
11	44				44	81.9	24.0	106.0	Normal pregnancy
12	34	1.7	3.5		60	82.5	14.5	97.0	Adenomatous thyroid, myocardial degeneration
13						86.0	11.0	97.0	Normal puerperium
Group average					58.2	74.7	17.3	92.0	

It may be noted that all cases in which there was a history of scarlet fever and rheumatism, and cases in which chronic tonsillitis had occurred, as well as many cases of pyorrhea and of six cases of gonorrheal infection are included in table 3. With few exceptions the results of phenolsulphonaphthalein and the sodium benzoate tests average higher in table 2 than in table 3. This difference, however,

TABLE 2

Normal cases

Case	Infection	Time	Phenolsulphonphthalein return	Sodium benzoate return		
				Two hours	Third hour	Total
2	Measles	1912		105	15.5	121
	Influenza	1918	40	(89)	(15)	(104) ²
	Gonorrhea	1922		74	15	89
	Postpartum*	7 weeks				
3	Diseases of childhood			86	11	97
	Appendectomy					
	Postpartum*	4 days				
4	Diseases of childhood					
	Influenza		58	81	17	98
	Tonsillitis	At 13 years				
	Postpartum	4 days				
7	Mumps in childhood (?)					
	Tonsillectomy	At 13 years	44	82	24	106
	Pregnancy	8 months				
14	In hospital on account of eyes	Until 9 years				
	Influenza	1918		72	32	104
	Hereditary syphilis (?)					
	Pregnancy	5 months				
8	Diseases of childhood		55	48	46	94
	Pregnancy	7 months				
20	Diseases of childhood					
	Tonsillitis	1914	50	64	31	95
	Influenza	1918				
	Pregnancy	8 months				
Average of 7 practically normal cases			49	75	25	100

* Sodium benzoate test.

TABLE 3
Abnormal cases

Case	Infection	Time	Phenolsulphonephthalein return	Sodium benzoate return		
				Two hours	Third hour	Total
			<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
1	Recent puerperal fever			25	47	72
	Postpartum*	2 weeks	43	(44)	(27)	(71)
	Postpartum*	3 5 weeks		62	7	69
5	Diseases of childhood		20			
	Influenza	1918	(31)	48	27	75
	Appendectomy		42			
	Gonorrhea					
	Term pregnancy*					
9	Diseases of childhood			37	15	52
	Tonsillitis		40	(54)	(14)	(68)
	"Rheumatism"			71	13	84
	Pregnancy	7 5 months				
	Pregnancy	8 months				
10	Appendectomy	1914	58	43	20	63
	Term pregnancy			(53)	(22)	(75)
	Postpartum	1 month		63	24	87
11	Pneumonia	At 11 years		42	17	59
	Influenza	1918	43	(47)	(13 5)	(61 5)
	Thyroidectomy	At 17 years		52	10	62
	Gonorrhea					
	Pregnancy	8 5 months				
	Postpartum	2 weeks				
12	Measles	1920				
	Scarlet fever		29	39	29	68
	Appendectomy	1917				
	Gonorrhea					
	Pregnancy	8 months				
13	Diseases of childhood					
	Scarlet fever		45	50	24	74
	Gonorrhea					
	Pregnancy	8 months				

* Sodium benzoate test

TABLE 3—Continued

Case	Infection	Time	Phenol-sulphonephthalein return	Sodium benzoate return		
				Two hours	Third hour	Total
			per cent	per cent	per cent	per cent
15	Diseases of childhood Measles Pneumonia Tonsillectomy Pregnancy	At 6 years 1921 8 5 months	43	53	30	83
16	"Rheumatism", measles Tonsillitis Term pregnancy		53	57	27	84
17	Diseases of childhood Scarlet fever, appendectomy Tonsillitis Pregnancy	1921-22 6 months	55	42	23	70
19	Acute rheumatism Tonsillectomy Term pregnancy	1921	27	64	22	86
18	Mumps Carbuncles Influenza "Toxemia of pregnancy" glycosuria Pregnancy	At 12 years 1916 1918 7 months	60	40	58 ²	93 ²
Average of abnormal cases			44	49	27	76

is considerably more definite in the benzoate than in the phenol-sulphonephthalein reactions. Grouping these cases according to their pregnant and postpartum condition gives no characteristic difference. In such a small series of cases with so many factors to consider, the effect of the pregnant state itself on the renal function as revealed by these tests is of course open to speculation.

Group 3. Subjects with essential renal involvement. In table 5 is presented a group of fifteen patients, all of whom show more or less

albumin and elevation, or recent history of elevation, of the blood urea at the time of the benzoate test. In four patients the blood urea was more than 100 mg for each 100 cc of blood and in one of these more than 300. All show considerable depression in the benzoate return, the highest three-hour return being 50 per cent instead of the normal 70 per cent. The eight patients showing most marked impairment (cases 1, 2, 3, 4, 5, 6, 8, and 9) also show a very marked and parallel depression of the reactions to both the phenolsulphone-

TABLE 4
Cases of pregnant and postpartum women (summary)

	Case	Phenolsulphonephthalein return	Sodium benzoate return		
			Two-hours	Third hour	Total
		<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
1 Average 7 "normal" cases		49	75	25	100
Pregnancy, 5 to 8 months	4				
Postpartum, 4 days to 7 weeks	3				
2 Average 12 "abnormal" cases		44	49	27	76
Pregnancy, 6 months to term	9				
Postpartum, 2 weeks, also 1 month	1				
Before and after delivery	2				
These showed improvement of 20 and 10 points in two-hour values, and of 24 and 3 points in third-hour values					
Difference		5	26	+2	24

phthalein and sodium benzoate test. The highest value for the former was 35 per cent, most of them being 25, 20, or lower, many even indicating no return. The highest two-hour benzoate return was 30 per cent, while the lowest reached the extremely low, yet definite amount of 4.3 per cent, or almost complete retention.

Of five patients (cases 7, 10, 11, 12, and 15) in this group having phenolsulphonephthalein values above 40 per cent, or within the normal limits of that test, four (cases 7, 10, 12, and 15) had more or

TABLE 5
Cases of rather marked renal lesions

Case	Milligrams for each 100 cc.		Uric acid	Non protein nitrogen	Phenolsulphonephthalein return	Sodium benzoate return			Diagnosis
	Urea	Creatinine				Two hour	Third hour	Total	
			per cent	per cent	per cent	per cent	per cent	per cent	
	Normal					70 0	20 0	90 0	
1 {	180	9 8	4 4		10 0	4 3	4 9	9 2	Glomerulonephritis, uremia
2	135								
	118	12 9	6 3		Trace	5 3	1 8	7 1	Left posterior nephrectomy right nephrolithiasis
3	320	16 0			0	6 9	7 4	14 4	Tuberculosis of ureter, kidney and bladder
					Faint trace				
4	293	11 3	4 8		Faint trace	7 6	6 4	14 0	Glomerulonephritis, uremia
5	161	3 2			30 0	13 0	14 8	27 8	Glomerulonephritis, uremia
									Exophthalmic goiter
6 {	90	4 2			20 0	14 0	13 8	27 8	Arteriosclerosis
	63								Pylonephrosis
7 {	96				50 0	20 1	14 2	34 3	Myocardial degeneration, en- dothelioma of bladder, car- cinoma of bladder
	80								
8	67	6 5	3 8	1 002- 1 010	10 0	21 7	11 0	32 7	Glomerulonephritis
9	67	4 3	5 5		15 0	30 0	17 2	47 2	Glomerulonephritis, malignant hypertension
10	43	1 5		4 6	45 0*	30 5	25 8	56 3	Polycystic kidneys, postop- erative
11	32	1 5	3 1		40 0	43 3	15 5	58 8	Exophthalmic goiter, general anasarca, cardiac decom- pensation
12	62	1 8	5 0	1 004- 1 019	55 0	43 3	17 7	61 0	Embolus nephritis
						53 6	17 8	71 4	
13 {	97	3 1	5 0	1 001-	30 0	49 6	30 6	80 2	Subacute glomerulonephritis
	57	1 7	3 4	1 017					
14 {	78	1 7	5 7		35 0	50 2	25 2	75 2	Chronic nephritis
	55	2 1							
15 {	52	1 6	11 0		55 0	50 5	27 4	77 9	Acute glomerulonephritis
	22	1 5	4 0						
Group average					26 8	26 3	15 6	41 9	

* Retarded at cystoscopic examination

less elevation of the blood urea. In each the benzoate return was definitely low for the two-hour period, which is in contrast to the phenolsulphonephthalein, but roughly parallel to the blood urea. It would seem that, while the patient's general and renal condition had undoubtedly improved, the benzoate test gave the more accurate conception of the degree of residual renal impairment.

Of the patients with nephrosis (table 6) all but one (case 5) showed slight but definite impairment of renal function by the criterion of the benzoate test. In two of these (cases 1 and 2) the benzoate return

TABLE 6
Cases of chronic nephrosis

Case	Milligrams for each 100 cc.		Uric acid	Phenolsulphonephthalein return	Sodium benzoate return			Diagnosis
	Urea	Creatinin			Two-hour	Third hour	Total	
			per cent	per cent	per cent	per cent	per cent	
	Normal				70 0	20 0	90 0	
1	13 0	1 1	3 7	45 70	59 0	20 4	79 4	Nephrosis
2	35 2	1 9	4 6	65 80	60 8	10 2	71 0	Nephrosis
3	58 0	2 0	3 8	35	63 4	15 6	79 0	Anemia, nephrosis (subacute glomerulonephritis)
	19 0	1 5						
4	22 0	1 3	2 2	65	64 6	31 9	96 5	Nephrosis
5	30 0	1 2	2 7	80	70 0	11 0	81 0	Nephrosis, tonsil sepsis
Group average				57	63 5	17 8	81 3	

appeared to be relatively more depressed than the phenolsulphonephthalein output and the blood urea would seem to indicate. It appears then that in nephrosis the benzoate output is relatively much higher than in other severe renal lesions, but follows the same general tendency as the urea and phenolsulphonephthalein. Thus it appears that in the human there is a condition analogous to that found by Kingsbury and Bell (23) in the rabbit, that is, that severe injury to the convoluted tubules does not greatly affect the synthesis of hippuric acid. The relatively high benzoate output in these nephroses

suggests that in the benzoate test we have a further aid in differentiating this type of case in which the damage is mainly tubular (parenchymal), and the type already considered, especially nephritis per se, in which the damage is mainly glomerular

The recent work of Snapper and Grunbaum (47) may be mentioned in this connection. They administered sodium benzoate in 5 gm doses in cases of various types and compared the elimination after the first 12 hours with that after the second 12 hours. They found that in cases of nephrosis and in those with arteriosclerotic kidneys as well as in normal persons, this dosage was, with the exception of a few hundred milligrams, eliminated quantitatively in the urine within twelve hours, provided there was no urea retention in the serum. In contrast to these cases they found that in similar patients with urea retention the elimination of sodium benzoate might require forty-eight hours, and they questioned whether in such cases there was a delay in the synthesis of hippuric acid or only a delay in its elimination. They inclined toward the latter view because, since hippuric acid is a nitrogenous substance, and since the contracted kidney has difficulty in secreting urine with a high concentration of nitrogenous substances, they believe there must be a close relation between water elimination and hippuric acid elimination. They were able to show that after the administration of sodium benzoate to patients with contracted kidneys, hippuric acid can be demonstrated in the blood in amounts up to 90 mg for each 250 cc of blood, whereas in patients with normal kidneys no hippuric acid, or only spurious traces of it, can be found under like conditions.

Snapper (45) was also successful in demonstrating that within twelve hours human beings with normal kidneys can eliminate hippuric acid that can be synthesized from 5 gm of sodium benzoate, regardless of any other affections. This is found to be true in various types of cancer and various types of cholelithiasis and gastric ulcer, in croupous pneumonia and also in cases with derangements of bile secretion, for instance, patients with cancer of the pancreas with complete icterus. Snapper likewise appreciated the possibility that when glycolic acid is diverted from the body hippuric acid synthesis might be retarded, thus he did not find to be the case. If these conditions occur with doses as large as 5 gm they certainly should occur

with a smaller dose of 2.4 gm. employed in the sodium benzoate test. Therefore, the work of Snapper et al. tends to strengthen our belief that in this test we have a proper means of estimating the ability of the kidney in synthesizing hippuric acid, and accordingly a reliable index to its general functional capacity.

Group 4 Subjects with only slight renal involvement The limits of this group were rather indefinite (table 7). These patients had more or less hypertension, more or less arteriosclerosis, more or less myocardial degeneration, but presented normal values of the blood

TABLE 7
Cases of hypertension

Case	Milligram for each 100 cc.		Uric acid	Non protein nitrogen	Blood pressure		Phenolsulphonephthalein return	Sodium benzoate return				Diagnosis
	Urea	Creatinin			Systolic	Diastolic		Two-hour	Third hour	Total		
			per cent	per cent	mm Hg	mm Hg	per cent	per cent	per cent	per cent		
1	Normal 21 0	1 5	5 3		210	130	45	70 0	20 0	90 0	Arteriosclerosis, hyper- tension, obesity Malignant hypertension Moderate hypertension, multiple adenoma, thyroid Essential hypertension	
2	32 0	1 7	3 7		220	140	65	68 1	19 3	87 4		
3	34 0	1 7		35 0	175	88		74 9	13 6	88 5		
4	38 0	1 4	3 2		208	113	60	82 5	14 5	97 0		
Group average							57 3	72 1	16 7	88 8		

urea and phenolsulphonephthalein return. They had few symptoms pointing to renal disease, and their renal function seemed still to be adequate to their present needs. In such cases there was moderate reduction in the benzoate output, but certainly less than in cases of actual nephritis.

The fact that in cases of this type (table 7, case 1), the benzoate return is reduced before the phenolsulphonephthalein return and blood urea retention, whereas in some of those in the preceding group (table 5, case 15) it is more tardy in returning to normal with improvement in the renal condition, warrants the following suggestion:

in early cases of renal damage in which the general excretory function of the kidney, as shown by other tests, is good, the evidence points to impaired hippuric acid elimination, later with progress of the lesion, the major role in the retention is usurped by the inability to synthesize the hippuric acid. This is, of course, of purely academic interest, and the fact apparently remains, that in this test there is an earlier indication of renal impairment than in our present interpretations of phenolsulphonephthalein excretion and blood urea retention.

It is in the border-line cases in this group in which, with advancing life and the gradual impairment of the various systems, a test indicating the "margin of renal safety" would be of greatest value. The desirability of having such a test is evidenced by the statement of a prominent pathologist (42) that probably 75 per cent of the functioning renal tissue is destroyed before the first clinical signs of renal insufficiency appear. The benzoate test may be more sensitive to these slight changes than other tests employed in this field.

Group 5 Cases with definite hepatic involvement It was realized that one of the principal objections, physiologically speaking, to the benzoate test of renal function, might be the influence of lowered hepatic function on the synthesis of hippuric acid. In order to determine the extent of this influence tests were made on a number of selected patients with disease of the liver, some of which exhibited not only clinical evidence of organic hepatic disease, but also functional deficiency as indicated by the tetrachlorophthalein and other tests. The degree of this functional impairment, as indicated by the retention of the dye in the blood-stream of these patients is noted in table 10.

All but three of the patients (cases 8, 9, and 10) had a rather definite reduction in the two-hour benzoate output. Five of them (cases 6, 7, 8, 9, and 10) had a three-hour value within 8 per cent of the normal (90 per cent). Even these patients, whose pathologic processes seemed most nearly limited to the liver, seemed to have impairment of the benzoate excretion in the first two hours, but the excretion tended to become more nearly normal in amount during the third hour. These findings are comparable to those of Delprat and Whipple (14), who noted delay in the output of hippuric acid in dogs with hepatic injury, it also illustrates a tendency, notable throughout this work, of the third-hour value to compensate for the two-hour value,

TABLE 8
Per cent of free benzoic acid excreted by patients

Case	Free benzoic acid			Difference*		Diagnosis
	Two hour	Third hour	Total	Two hour	Third hour	
	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	
1	0.72	1.86	2.58			Hypertension, obesity
2	0.62	0.30	0.92			Acute glomerulonephritis
3	0.75	0.36	1.11			Malignant hypertension
4	1.20	1.05	2.25			Nephrosis
5	0.83	1.34	2.17			Chronic glomerulonephritis
6	2.16	0.86	3.02	60.0	84.1	Biliary cirrhosis
7	1.05	1.24	2.29			Chronic nephritis
8	1.56	1.04	2.60			Emboic nephritis
9	1.11	0.67	1.78			Nephrosis
10	0.30	0.21	0.51	3.9	8.7	Chronic glomerulonephritis
11	3.23	2.87	6.10	49.2	80.7	Biliary cirrhosis
12	3.41	2.21	5.62	48.8	65.4	Common duct stone, obstructive jaundice
13	2.87	3.36	6.23	35.9	68.8	Infective jaundice
14	0.53	0.87	1.40			Tuberculosis of the kidneys
15	0.86	0.69	1.55			Carcinoma of the bladder, pyelitis
16	1.56	1.48	3.04	82.5	91	Stricture, common bile duct
17	1.45	1.66	3.11	25.1	35.7	Obstructive jaundice (postoperative)
18	4.10	3.80	7.90	68.1	76.7	Hypertension, gallbladder
19	0.69	0.57	1.26	4.6	5.8	Left, nephritis, right, nephrolithiasis
20	1.49	0.78	2.27	6.1	11.7	Chronic glomerulonephritis

* The difference is between the total and free benzoic acid values, that is, the value for the conjugated, non-toxic hippuric acid expressed as benzoic acid in percentage terms of the administered dose

TABLE 9
Comparison of group averages

	Type	Cases	Phenolsulpho- naphthalein return	Sodium benzoate return		
				Two hour	Third hour	Total
				<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
			<i>per cent</i>	70.0	20.0	90.0
Table 1	Normal	13	58.2	74.7	17.3	92.0
Table 6	Hypertension	4	57.3	72.1	16.7	88.8
Table 7	Nephrosis	5	57.0	63.5	17.8	81.3
Table 12	Hepatic	10	53.0	49.4	22.4	71.8
			(3 cases)			
Table 5	Nephritis	15	26.8	26.3	15.6	41.9

thus making the total three-hour value approach normal. This probably indicates a tendency of the synthesis and elimination to proceed at a maximum rate, hence the influence of diet may not be great, as the damaged mechanism can (?) only eliminate so much hippuric acid for each unit of time, regardless of the amount which may have accumulated in the blood stream.

TABLE 10

Degree of functional impairment of liver, and summary of laboratory findings in cases of rather marked hepatic involvement

Case	Milli grams for each 100 cc		Uric acid	Non protein nitrogen	Tetrachlorophthalic retention	Phenolphthalein return	Sodium benzoate return			Grade*	Diagnosis
	Urea	Creatinin					Two-hour	Third hour	Total		
			per cent	per cent		per cent	per cent	per cent	per cent		
Normal							70.0	20.0	90.0		
1	35			29	4		23.4	27.1	50.5	3	Toxic jaundice after arsenphenamine treatment
2							26.5	12.3	39.3	3	Obstructive jaundice
3	26.1	1.1	1.8		4		38.7	36.3	75.0	3	Jaundice 3
4	28	1.4	3.1		2		26.5	24.1	30.6	4	Carcinoma of liver?
	15				2		51.2	15.5	66.7	2	
5	36		36.5		4		52.3	18.7	71.0	2	Obstructive jaundice
6	10				4	40	52.4	34.4	86.7	2	Biliary cirrhosis
7	10.1	2.2	2.0		3	80	58.5	23.9	82.4	2	Cirrhosis
8	22				4		62.2	24.9	87.1	1	Cirrhosis
9	29.1	6.1	1.6		3	45	64.8	20.7	85.5	1	Cirrhosis
10	12.1	4.1	1.7		1		84.1	9.9	94.0		Postoperative stricture of common duct
Group average						53	49.4	22.4	71.8	2	

* See table 11

In order to obtain a basis of comparison in this group between the two tests of greatest significance, the benzoate elimination values for the two-hour period are expressed by grades (see table 11). On this basis table 12 is obtained from table 10.

In this comparison it appears that in the first four cases there was a somewhat parallel retardation in the elimination of the two sub-

stances, and in the remainder the lack of parallelism was seemingly the prominent feature, in one (case 8) the dye being retained to a marked degree, while the benzoate excretion closely approached normal. In none of these cases was there any notable retention of urea in the blood, which would suggest that the kidneys were functioning normally, and that any impairment in hippuric acid excretion

TABLE 11

Grades	Elimination*
	<i>per cent</i>
1	60-70
2	50-60
3	20-50
4	Below 20

* Normal elimination is 70 per cent or more

TABLE 12

Comparison of tetrachlorophthalein and benzoate elimination in ten cases

Grade	Tetrachlorophthalein elimination grade	Benzoate elimination, grade
1	4	3
2	—	3
3	2	3
4	{ 4	{ 4
	{ 2	{ 2
5	4	2
6	4	2
7	3	2
8	4	1
9	3	1
10	1	1
Average	3	2

was due to other causes, presumably hepatic changes. Before accepting such a hypothesis one must remember that in jaundiced patients, as was notably true for the first four patients in this group, the values for blood urea are decreased (4).

The possibility of hepatic damage limiting hippuric acid synthesis is by no means excluded, in fact, the view is well supported that hepatic damage does delay it. Viewing this group broadly, the

evidence would seem to demonstrate impairment of renal function rather than hepatic disease *per se*. Such a view would be in harmony with the work of the recent group studying hepatic function at the Mayo Clinic, especially Counsellor, who has histologically demonstrated definite lesions of the nephrosis type in patients who have been suffering from rather protracted obstructive jaundice. With only two of ten three-hour values for benzoic acid in table 10 below 66 per cent, whereas in table 5 only four of fifteen were above it, it appears that in this group there is little evidence of extensive renal damage, even by the criteria of the benzoate test, certainly there is definite contrast between the two groups.

In this connection Snapper's work should be recalled, which showed that patients with complete obstruction of the common duct and those with complete biliary fistula did not differ from those of normal persons in their ability to synthesize and eliminate hippuric acid. However, the twelve-hour test period which he employed would not have revealed any delay in the first two hours as our experiments suggest, as do also those of Delprat and Whipple (14), and of Lachner, Levinson, and Morse (30). Morgulis' (39) suggestion that the liver may play a subsidiary rôle in the synthesis in the mobilization of glycogen is strengthened by our finding a much greater amount of free benzoic acid in this group than in any other.

EXPERIMENTS WITH DOGS

Dogs were selected whose livers had been subjected to experimental injury of various types. Each received a dose of 0.5 gm. of sodium benzoate for each kilogram of body weight, administered intravenously in a 20 per cent solution. Only female dogs were used and specimens of urine were obtained by catheterization two, five, ten, and twenty-four hours after injection of the benzoate. These specimens were examined for total and free benzoic acid by the methods described. In table 13 the benzoic acid return is indicated in terms of the percentage of the administered dose. In the two normal dogs there is similarity not only in the returns for the shorter periods but in the differences between the total and free benzoic acid values, indicating that the synthesized benzoate or hippuric acid is almost identical, 53.4 and 51.8 per cent.

TABLE 13
Summary of experimental work on dogs

Dog	Renal function				Hepatic function					Sodium benzoate return					Diagnosis
	Albumen	Phenolsulphonethalein return per cent	Urea mg	Non protein nitrogen per cent	Bilirubin	Levulose	Calcium time minutes	Coagulation time minutes	Tetrachlorophthalein retention	Two hour per cent	Five hour per cent	Ten hour per cent	Twenty four hour per cent	Total per cent	
H27 1	1	60	15	32.7	0	0	3.0	4	0	7.6 0.98	14.2 2.5	4.6 0.79	33.0 1.84	59.5 53.4 6.13	Normal
A633 2										9.7 4.0	10.1 1.9		38.0 0.97	57.8 51.8 6.0	Normal
G926 3 5-8-24	2	80	10	18.2			7.5	7	4	22.8 6.6	15.6 0.73	8.7 2.9	26.8 12.6	73.9 51.1 22.8	Obstructive jaundice
5-21-24										5.7 3.5	13.1 2.1				Obstructive jaundice
5-28-24	1	70	51	25.5			5.0	5	2	5.2	15.2 2.8	21.2 ?	33.2 0.29	75.8	Obstructive jaundice

11121 1 6-15-21	Trace		34	51 2	0	0	0	5 0	7	0		16 6 0 13	14 2 6 2	22 1 13 6 2 7	38 7 ?	91 6	6 cc chloroform
6-17-24	2		12			0	0	8 5	6	3							Chloroform injected
11108 5	1	60	26	32 6				4 5	5	3		10 3 ?	16 5 6 17		51 3 2 1	78 1 12 8?	1 experiment cirrhosis
G592 6 6-12-21												1 3 1 5	10 7 0 38		50 8 2 2	65 8 11 0?	Eck fistula
6-17-21												3 03 1 12					Hepatectomy

Dog 3 with definite, severe, and progressive obstructive jaundice was given benzoate on three occasions. The phenol-tetrachlor-phthalein and bilirubin tests indicated definite hepatic injury. The values for the benzoate determinations, however, were, at the time of the first examination, very nearly parallel to those of the normal dogs, the difference being 51.1 per cent, the values for the shorter periods did not vary more than the corresponding ones might for normal dogs. Later examinations showed no significant change from the first examination. Here then, despite a progressive hepatic lesion the benzoate values were essentially normal, as were the renal function tests.

Dog 4 had normal renal function, normal phenol-tetrachlor-phthalein return, normal coagulation time, and no serum bilirubin. The following day 6 cc of chloroform in oil was given intramuscularly. A day later there was bilirubin in the serum, an increased coagulation time, and marked retention (grade 3) of phenol-tetrachlor-phthalein; these results all point to definite acute impairment of hepatic function. In this case the amount of synthesized benzoate at the end of two hours was 16 per cent, and at the end of ten hours 41.6 per cent, values essentially equivalent (?) to those found in the normal dogs.

Dog 5 had six intra-portal injections of chloroform, at operation just preceding the injection of benzoate the liver was seen to be shrunken and distorted by bands of fibrous tissue. The liver function tests showed considerable serum bilirubin and notable (grade 3) retention of phenol-tetrachlor-phthalein, the renal function, on the other hand, was normal as shown by the phenol-sulphone-phthalein test (60 per cent). Despite the hepatic injury the synthesized benzoate values appear quite normal, 65.3 per cent being synthesized during the twenty-four hours following the administration of the benzoate, and the values for the shorter periods were quite comparable to those found in normal dogs.

Dog 6, when first examined, had an Eck fistula, made preparatory to hepatectomy, while no functional tests had been made it is known that the hepatic function in such cases is somewhat impaired. The values following the first benzoate administration were essentially normal. Somewhat later this dog was hepatectomized by Mann (37) by his own method, but unfortunately the dog became anuric

after the collection of the two-hour specimen. This specimen showed a definite synthesis of hippuric acid although the amount was smaller than on the previous examination. This may have been due to the factors causing the anuria, such as fall in blood pressure or surgical shock, rather than to failure of the detoxicating mechanism itself. The absence of the liver may have been responsible for the anuria, since with the failure of glycocoll mobilization, which we conceive as the hepatic rôle in this synthesis, the rather large dose of sodium benzoate became actually toxic. It would be interesting to know whether synthesis might not have taken place essentially as well in such a dog as in others had glycocoll been given intravenously following the hepatectomy.

This work on dogs strongly supports the belief that in the dog the liver has no direct concern in the detoxication of hippuric acid, but that it is probably essential to mobilization of the glycocoll necessary to the synthesis. This work seems in harmony with the results noted in human patients, although it is not directly comparable since in the dog benzoic acid is not normally conjugated to hippuric acid to as large an extent as in the human.

FREE BENZOIC ACID

Morgulis and his co-workers objected to the benzoate test as proposed by Kingsbury on the basis of inaccuracies due to the fact that a certain amount of benzoic acid is excreted in the free state. In table 8 our findings in this regard are abstracted. Two-hour and three-hour values for free benzoic acid were obtained in twenty of the cases (table 8). The per cent of free benzoic acid, relative to the administered dose, is indicated and the values are therefore comparable to those of the total benzoic acid eliminated. The results may be summarized as follows: two cases 0 to 1 per cent, five cases, 1 to 2 per cent, six cases, 2 to 3 per cent, and seven cases more than 3 per cent.

It is significant that all of the patients with values above 3 per cent had hepatic impairment, of these, two had biliary cirrhosis, and two obstructive jaundice, one was a case of infectious jaundice, one had a stricture of the common bile duct and the last had cholecystitis.

This evidence gives weight to the hypothesis of Morgulis that the rôle of the liver in this particular function is secondary only and is mainly concerned with the formation or mobilization of glycocoll. Such an hypothesis would lead one to suppose that there is, at a given time, insufficient glycocoll in the blood stream for a maximal detoxication by the kidneys, which, being intact, detoxicate and eliminate the benzoic acid as fast as the requisite glycocoll is brought to them. This would be in harmony with the situation described in this section of our work, with the delay observed by Delprat and Whipple, and by Lachner, Levinson, and Morse, with the increased formation of hippuric acid with increased glycocoll feeding as described by such workers as Lewinski, Griffith and Lewis, and Csonka, and with the delay in hippuric acid elimination observed by us in hepatic cases during the first two-hour period of the test, tending to attain by the end of the third hour a normal full three-hour value. In the hepatic cases there appeared to be a faulty synthesis rather than the faulty elimination observed in the severe nephritic cases.

The error of this test as an indicator of renal function is probably greatest in cases with hepatic disturbance, in such cases the three-hour total value is most significant from a renal standpoint. From our data it appears that a three-hour free-benzoic acid value which is above 3 per cent after giving 2.4 gm. of sodium benzoate is highly suggestive of some hepatic disturbance. On the other hand, from table 8 it appears that even in cases of marked renal insufficiency (cases 10, 19, and 20) much smaller amounts of free benzoic acid are excreted in the urine and at no time were these great enough to detract from the clinical information available from the test.

DISCUSSION

Since the main points indicated by the examination of the various groups were discussed in their appropriate relations, little in the nature of a general discussion is necessary. A few points will, however, be emphasized.

The synthesis of hippuric acid is not seriously interfered with by diseases of the parenchyma of the kidney, as severe nephrosis. In destructive lesions of the kidney, such as those due to tumors, multiple

cysts, tuberculosis, or nephritis, there is marked interference with its elimination. Although, according to the work of Snapper, it appears that even here the hippuric acid may be synthesized and merely dammed back in the blood stream, this seems to us decidedly open to doubt in the severest cases.

The two-hour value, from a renal standpoint, is probably the more significant figure of the test for reasons heretofore elaborated. In this connection a suggestion might be offered for the use of the benzoate test in conjunction with the phenol-sulphone-phthalein test. The diet had best be controlled with regard to fruits, especially cranberries, for from twelve to twenty-four hours preceding the test. Immediately after voiding the morning specimen, the usual amount of phenol-sulphone-phthalein should be given intravenously. Fifteen minutes later the patient should take 2.4 gm. of sodium benzoate as described. The urine for two hours and fifteen minutes should be collected as a single specimen, carefully measured, and several portions taken for the determination of the phenol-sulphone-phthalein and total benzoic acid values.

CONCLUSIONS

1 The sodium benzoate test is of value in determining the functional capacity of the kidneys. No other type of affection so definitely and markedly affects its synthesis or elimination. However, a free benzoic acid value above 3 per cent of the administered dose of sodium benzoate suggests a secondary hepatic lesion, the high value probably being due to the inability of the liver to mobilize glycocholic acid, it probably should not be construed as reflecting against the ability of the kidney to detoxicate the substance. In other cases the amount of free benzoic acid is probably negligible, clinically speaking.

2 The benzoate test is probably a more delicate indicator of early functional renal disturbance than other tests in common use, and, therefore, should be of greatest value in patients with early suspected renal involvement or in patients whose phenol-sulphone-phthalein or blood urea tests are still normal or have returned to normal. The test has no particular advantage over other tests in advanced cases, inasmuch as the technique is more time consuming, although this

factor, in a well organized laboratory in an institution where many renal cases are studied is by no means prohibitive

3 The results of these experiments confirm those of Kingsbury regarding the standard of "normal" excretion for this test, which is an excretion of 70 per cent or more of the benzoic acid equivalent of the sodium benzoate in two hours, and from 85 to 90 per cent or more, in three hours

4 The work on dogs gave little evidence that the ability of the animal to synthesize and eliminate hippuric acid was decreased when the liver was damaged or absent, except that there was a lack of available glycocoll, making the synthesis impossible for other organs, such as the kidney

5 Further work might demonstrate the determination of free benzoic acid elimination, after administration of a standard dose of sodium benzoate, to be of value as a functional test in establishing the degree of hepatic impairment

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STUDIES ON THE MORNING ALKALINE TIDE OF URINE IN NORMAL PERSONS AND IN PATIENTS WITH NEPHRITIS¹

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(Received for publication June 22, 1925)

INTRODUCTION

The hydrogen-ion concentration of the urine has been the subject of much interesting study, by both pathologist and clinician, especially since the work of Henderson and Palmer. These investigators, using twenty-four hour specimens, studied the hydrogen-ion concentration of the urine both in normal persons and in patients with various diseases, including nephritis. They found that on the average the urine of nephritics was definitely more acid than that of normal persons. It has long been well-known that in normal individuals the urine is more alkaline during the morning. In 1919, Leathes investigated the changes that took place in the morning specimens in cases of acute and subacute nephritis in soldiers, and found that the alkaline tide, which was always normally present, was sometimes reduced or even absent in nephritis.

As there were such good opportunities at the Mayo Clinic to study cases of nephritis, it seemed important to discover what bearing Leathes' test might have on treatment or prognosis. Studies were accordingly undertaken with this object in view.

METHODS

The first series of cases was investigated according to a modification of Leathes' technic (table 1). His method of demonstrating the alkaline tide was as follows:

¹ Submitted to the Faculty of the Graduate School of the University of Minnesota in partial fulfillment of the requirements for the degree of Master of Science in Medicine, March, 1923.

A night specimen of urine was collected at 7 00 a m for the period from 11 00 p m to 7 00 a m , and the second specimen at 8 00 a m , immediately after which 500 cc of water was given. Hourly specimens were then collected until twelve o'clock. No other fluid or food was taken by the patient after the evening meal preceding the test until its completion. By using suitable indicators, and titrating with tenth-normal acid and tenth-normal alkali, Leathes estimated the alkalinity per cent of the urine. Henderson and Palmer had previously described a similar method for obtaining the ratio of actual to total possible alkali excreted, by titrating with acid and alkali to a known hydrogen-ion concentration. It will be noted that by giving 500 cc of water the ability of the patient to excrete water could be tested, and the range of specific gravity determined. In all the cases described the amount and specific gravity of each specimen was noted.

A series of buffer solutions were made up according to the method of Henderson and Palmer. At a later date these solutions were made up from Clark's tables. The accuracy of the pH solutions was checked by means of the electro-titration apparatus of Wendt, and in some cases the pH of the urine was estimated by this means as well as by the colorimetric method. The pH was estimated in all cases except those in table 1.

In the course of the work it became apparent that the hydrogen-ion concentration of the urine may change soon after it is voided. It has been shown by Gamble, and by Marshall, that on standing or shaking, carbon dioxide is liberated from the urine, and that the resulting change in pH may be quite marked, especially in the more alkaline urines. It was therefore decided to run a series of urine samples from normal and abnormal cases, estimating the carbon dioxide and phosphates in addition to the properties mentioned. The phosphates were of particular interest as they are largely responsible for the buffer action of the urine.

The carbon dioxide in the various specimens was estimated by van Slyke's method, and the phosphates by Doisy and Bell's colorimetric method. In this group of cases, all males, every precaution was taken to avoid any loss of carbon dioxide, and the consequent change in hydrogen-ion concentration. The urine in each case was passed by means of a funnel which emptied below a layer of toluene into an especially prepared narrow cylinder. This was at once corked, and, care being taken to avoid any shaking, carried to the laboratory in a neighboring room, where the carbon dioxide was at once estimated and the pH determined, immediately after this the alkalinity per cent was estimated. The complete study as outlined was carried out in all the normal cases (table 2) and in certain of the abnormal cases included in tables 3 and 4. The carbon dioxide and phosphate results for both normal and abnormal cases are shown in table 4.

One of the simplest tests for changes in the acid-base equilibrium in the individual was the direct determination of the alkali reserve of the blood plasma by van Slyke's method. It seemed important to determine at the onset whether there were not definite abnormalities in the excretion of acid by the kidneys, with little or no decrease in the carbon dioxide combining power of the blood plasma. Accord-

ingly, in a group of both normal and abnormal cases, I determined this factor, using samples of blood obtained both before water was given and at the time the diuresis should be at its height (table 7)

RESULTS IN NORMAL CASES

The minimal excretion from 8 00 a.m. to 12 00 m., after 500 cc of water had been given at 8 00 a.m., was 260 cc, the maximal 1066 cc, and the average 699 cc. The minimal specific gravity at the height of diuresis varied from 1 003 to 1 012, while the maximal varied from 1 024 to 1 035. The smallest range difference was 0 020 in a case in which the specific gravity varied from 1 012 to 1 032. The greatest range difference was 0 030, the specific gravity varying from 1 005 to 1 035. The average range difference was between 0 024 and 0 025. The initial alkalinity per cent varied from 21 to 56, and the maximal from 64 to 96. In every case there was a definite and unmistakable rise in the alkalinity per cent during the morning, under the conditions of this test. The initial pH ranged from 5 35 to 6 2, with an average of 5 65, the maximal pH from 5 9 to 7 7, the average being 6 95. The phosphates always showed a decrease during the morning, the night specimen always containing the maximal amount. Following the early initial decrease there was a definite though slight increase later in the morning, even though the urine became more alkaline.

The results in these normal cases are in accord with those of Leathes, showing in every case a definite diuresis and a definite alkaline tide during the morning. Leathes also performed the test with varying amounts of water, even less than 500 cc, and found that the change in the alkalinity per cent still persisted. In one normal case I tried the test without administering water, and obtained a normal curve for the alkalinity per cent, and an increase in the pH figures as in the other normal cases in which water was given.

The change in the pH toward the alkaline side during the morning showed a definite but not exact parallelism to the curve of the alkalinity per cent. The carbon dioxide increased relatively with the increase in alkalinity. In every case it was found that a higher pH figure, that is, a change to the alkaline side, was accompanied by relatively increased carbon dioxide, and correspondingly an increased acidity was always accompanied by a fall in the carbon dioxide. In other words

the carbon dioxid always varied directly with the figure of the hydrogen-ion concentration This is in accord with the work of Gamble and Marshall The total carbon dioxid naturally does not follow the pH so strikingly The decrease in the phosphates during the morning and the later increase correspond to the findings of Fiske

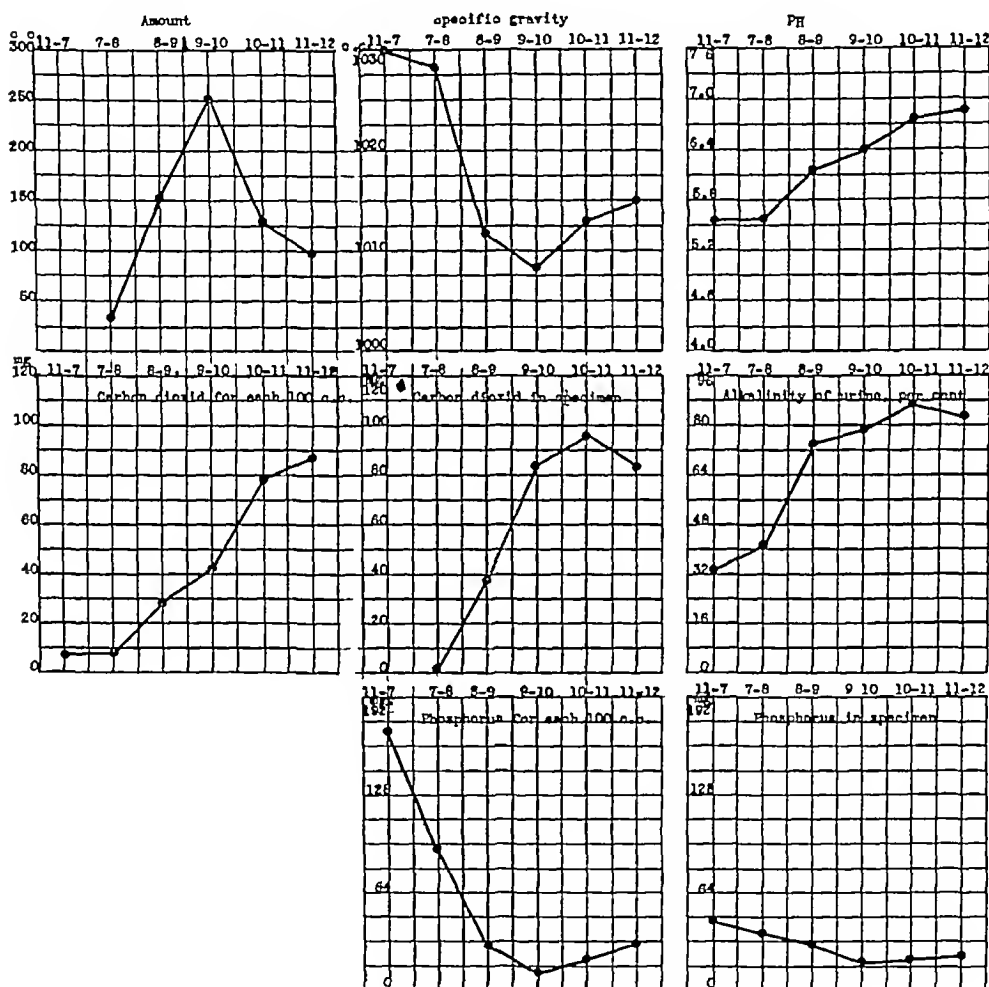


FIG 1 AVERAGE CURVES FOR TEN NORMAL CASES

The average curves of the various factors determined in normal cases are shown graphically in figure 1

RESULTS IN ABNORMAL CASES (TABLES 1, 3 AND 4)

Similar types of cases are grouped together and those in which the complete study was made are indicated From a study of these

tables it will be observed that, while in some cases the test shows that all the factors are within normal limits, in others there are marked deviations from the normal. Leathes divided the cases which reacted abnormally into three groups: (1) those with little or no diuresis, but a normal alkaline tide, (2) those with diuresis, but no alkaline tide, and (3) those with neither a diuresis nor an alkaline tide.

The alkalinity per cent findings which are corroborated by the pH figures show that most of the cases in this series belong in one or another of these groups, although there are very few in group 1. To these three groups of cases, however, I would add a fourth small group, in which there was a definite acid tide during the morning, a reversal of the normal alkaline tide. This group will be discussed later at greater length.

It is to be noted that, even though the urine may react fairly normally with regard to diuresis, alkaline tide, or both, the range of the specific gravity may still be very limited.

The carbon dioxide in all cases, as was to be expected, varied inversely as the acidity.

In cases without an alkaline tide, there is a tendency, which may be very marked, to a fixation of the amount of phosphate excreted each hour. A study of the tables showing the phosphate estimation in the night specimens of normal and abnormal cases brings out an interesting fact. Under the conditions of this test the maximal night concentration of phosphates in abnormal cases, estimated as milligrams of phosphorus for each 100 cc., is less than the minimal normal night concentration. The average figure for the abnormal cases likewise was much below the normal average. The maximal figure of total phosphates for the abnormal cases was the same as for the normals, but this was very exceptional, as the next highest figure, 244.9 mg., is almost the same as the minimal figure for the normals. On the whole, in abnormal cases, the phosphates, both total and relative, in the night specimen were definitely below normal.

DISCUSSION

In studying renal function from a new viewpoint, it is, of course, necessary that the results obtained be correlated with previously accepted standards. In this study seventy-four hospital cases were

investigated, the majority of which were suffering from definite renal disease, although a few other conditions were included. In tables 5 and 6 the main clinical and functional findings are correlated together with the results of the present studies with regard to the morning alkaline tide.

The tendency toward a lack of diuretic response is evident, although in some cases the response is well within normal limits. In comparison with the minimal normal excretion of water, 260 cc, it will be noticed that in a large number of cases this function is limited, sometimes very markedly. The marked tendency toward fixation of the specific gravity in the urine of many of the cases is very striking, especially in table 1.

A study of the alkalinity per cent figures in tables 1 and 3 will show in what a large proportion of cases of chronic glomerulonephritis there is an abolition, either partial or absolute, of the alkaline tide. To be well under the standard set by normal cases, I have considered that all with a difference between the initial and maximal alkalinity per cent of less than 15 should be considered in this class. In figure 2 a number of curves of the cases of this group are shown with the normal curve superimposed. The flattening of the curves is at once apparent.

Very few cases other than those of chronic glomerulonephritis and subacute diffuse nephritis fall below this standard, the exceptions including two cases of malignant hypertension, two of arteriosclerosis, two of focal nephritis, one of gout, one of bilateral polycystic kidneys, and one of bilateral hydronephrosis. This last case, in which there was an acid tide, is discussed with that group.

Figure 3 represents the curves found in a case of chronic glomerulonephritis which was clinically severe (case 49), the patient has since died. In each section of this figure the normal average curve is also given for comparison. As will be observed, the hourly output of urine is almost constant, the specific gravity is virtually fixed, and the alkalinity per cent shows very little change. The pH figure is the same for each specimen and is at a high acid level for urine. All these indicate a fixed acid urine. The carbon dioxide, in accordance with the pH, remains very low and fixed, and the phosphates also show a marked tendency toward fixation.

Thus it would appear that in such cases of chronic and subacute

glomerulonephritis the kidneys excrete a more or less fixed urine with the amounts of the main elements little changed from hour to hour. These facts indicate that the kidneys in such cases constantly excrete their maximal amount and have lost that reserve excretory function so characteristic of normal kidneys. The kidneys in this case (case 49) were unable, even at the height of their function, to eliminate the normal waste products, which therefore accumulated in the blood, as

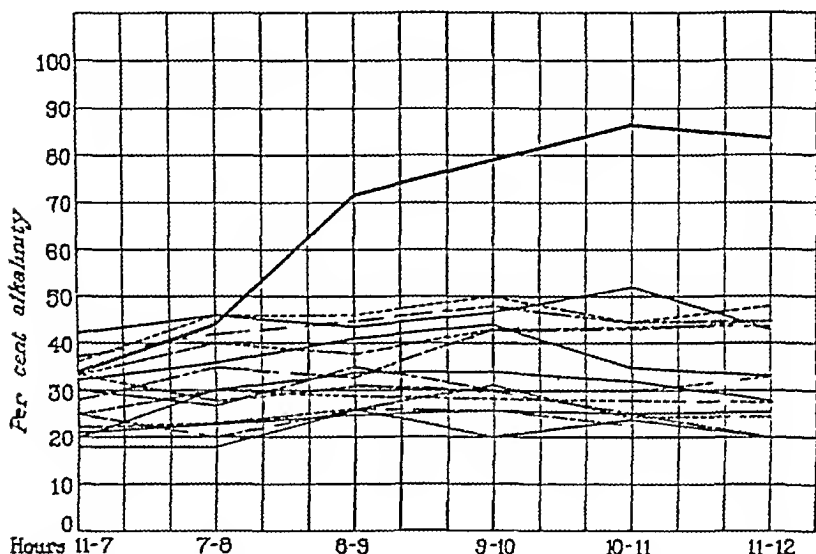


FIG 2 CURVES IN CASES WITHOUT ALKALINE TIDE CONTRASTED WITH CURVE IN CASE WITH ALKALINE TIDE

———— Composite normal Other curves, cases without alkaline tide

indicated by a blood urea of 345 mg for each 100 cc, and a blood creatinin of over 12 mg for each 100 cc

By studying the clinical findings given in tables 5 and 6, and by observing the number of patients in groups 1 and 3 who have died, an indication of the severity of some of the cases will be obtained. Included in these groups of course are other patients in whom the nephritis was clinically comparatively mild. Although the urinary findings obtained in different cases by this test vary considerably, as would be expected in general it may be said that only in cases of

clinically severe nephritis is there an abolition of the alkaline tide. These cases also show that there may be marked fixation of the specific gravity in spite of an alkaline tide, a diuresis well within normal limits, or both. As indicated in the results with abnormal cases,

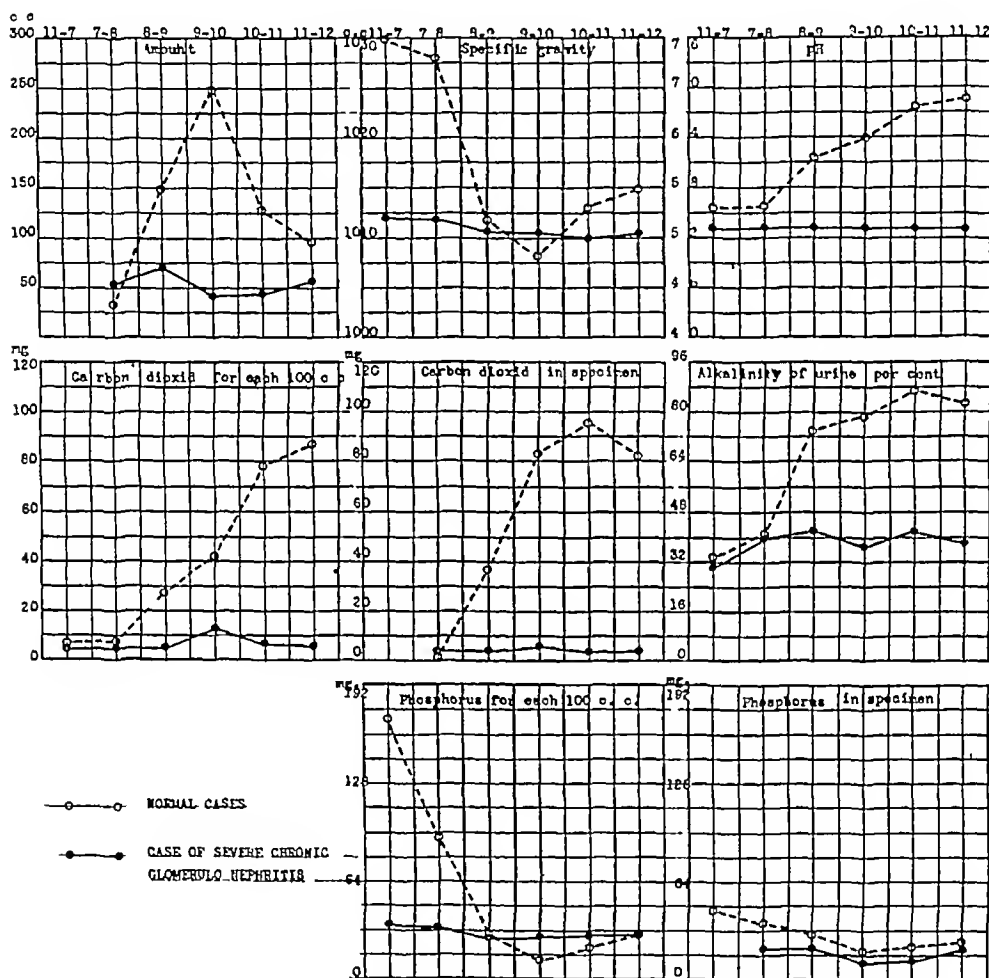


FIG 3 CURVES IN CASE OF SEVERE CHRONIC GLOMERULONEPHRITIS CONTRASTED WITH CURVES IN AVERAGE NORMAL CASE

○ - - - - ○ Normal cases ● - - - - ● Case of severe chronic glomerulonephritis

some of the patients who were tested (besides those already mentioned) showed limitation of function in some respects, but these findings were not constant or marked enough to require particular comment.

Since the carbon dioxide in the urine depends on the acidity, it would

seem that the estimation of the carbon dioxide had no practical value. The tendency of the phosphates to be fixed in the cases with no alkaline tide is significant, but the lowering of the phosphate content of the night urine in cases with renal involvement would appear to be even more so. I have never seen any record of such a lowering of the phosphate content in the night urine of nephritic patients, and such an estimation may prove of practical value. However, further investigation along this line must be carried out. Variations may depend on the diet and on the partition of the phosphate excretion between urine and feces. All the abnormal patients in which this was found were on a limited, controlled diet, while the normal subjects were not restricted.

Table 7 gives the results of the estimation of the carbon dioxide combining power of the blood plasma in a number of normal and abnormal cases. In none of the abnormal cases studied in regard to this point were there symptoms suggesting acidosis. As was to be expected, the alkali reserve of the blood of normal subjects showed no significant change during diuresis. In the group of clinical cases of nephritis studied, there was no diminution of the reserve alkalinity of the blood, and, as in the normal cases, there was no significant change during diuresis.

An acid tide was encountered in four cases, and in these there was a definite increase in the acidity during the morning. The first case in which this phenomenon was encountered was case 10. The result was thought at first to be incorrect, but the test was repeated three times without variation in the result. It is true that in this case the specimens were not examined as soon as they were passed, but, as this acid tide was an isolated phenomenon among the large group of cases and as it occurred so persistently, its presence may be considered as established. This case was one of chronic glomerulonephritis. The clinical findings were blood pressure 150 systolic and 90 diastolic, albumin 2, no casts or red blood cells, blood urea 289 mg. for each 100 cc., blood creatinin 6.2 mg. for each 100 cc., no edema, normal eye-grounds and a phenolsulphonephthalein return of 5 per cent. The patient died later.

This acid tide was next observed in case 70. The diagnosis, bilateral hydronephrosis, was confirmed at necropsy. The albumin varied

from 1 to 3 while the patient was in the hospital, the maximal blood urea was 327 mg for each 100 cc, the maximal blood creatinin was 22.6 mg for each 100 cc, edema was present, the eye-grounds showed a pallor of the discs but were otherwise negative, there was no return of phenolsulphonephthalein, and the blood pressure was 158 systolic and 94 diastolic. The pH in this and the remaining two cases confirmed the presence of an acid tide.

The third patient (case 40), was a young man, aged twenty-three, with a diagnosis of chronic glomerulonephritis. His urine contained albumin varying from 1 to 3, and microscopically showed a few casts, and a considerable number of red blood cells. The blood urea was 74 mg for each 100 cc, and the blood creatinin 1.8 mg for each 100 cc, edema was present, the eye-grounds were normal, the phenolsulphonephthalein return was 40 per cent, and the blood pressure 194 systolic and 122 diastolic. This patient felt well enough to be up and about. His death occurred shortly after leaving the hospital.

In the fourth case (case 46) the disease diagnosed chronic glomerulonephritis. His blood pressure was 200 systolic and 140 diastolic, his urine showed albumin 1 to 2, a few casts and red blood cells. His blood urea was 154 mg for each 100 cc, and his blood creatinin 2.7 mg for each 100 cc. There was slight edema. The retinal arteries showed a reduction of caliber, and there was a suggestion of edema of the discs with a small, diffuse hemorrhage above the left one. The phenolsulphonephthalein return was 5 per cent. This patient had a long and severe illness while here, his condition being considered very critical. The patient lived a short time on a reduced diet, and with limited activities. Following indiscretions he rapidly became worse, and died fourteen months after this test was made.

In this case each specimen was examined at once, and every precaution taken to avoid any change in hydrogen-ion concentration.

A summary of the findings in these four cases will be found in table 8 and figure 4, which gives the curves of the alkalinity per cent figures, the normal curve being superimposed for comparison. They show graphically how little variation there is in the hourly output of urine, the virtually fixed specific gravity, and the unmistakable acid tide. The crossing that occurs between the normal curve and those of the acid tide cases is also striking. The abnormal urine shows first a

relatively high alkalinity but becomes progressively less alkaline until noon, while in normal urine, the reverse is true

So far I have been unable to find any record of an actual demonstration of an acid tide in the urine. This occurred only in severe cases, and in three of the four cases bacterial infection of the lower urinary tract could be excluded. No alkaline treatment was being employed. I believe, therefore, that such an acid tide occurs only in the end stages of some cases with marked involvement of the renal parenchyma. Thus it would seem, no matter what the apparent

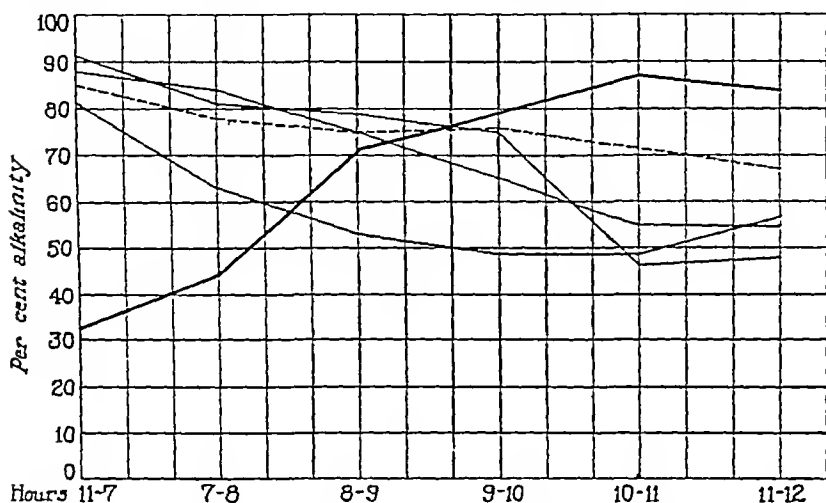


FIG 4 CURVES IN ACID TIDE CASES COMPARED WITH CURVE IN NORMAL CASE

———— Composite normal Other curves, cases with acid tide

general condition of the patient at the time of the test, that the presence of an acid tide is of grave prognostic import

It is suggested that a test similar to Leathes' test might with advantage be used instead of the water test of Volhard and Fahr, for the former shows not only the ability of the kidney to excrete water, and the range of specific gravity, but also the presence or absence of the normal alkaline tide. Any objection to a titration test could be overcome by substituting for the estimation of the alkalinity per cent the determination of the pH as done by Henderson and Palmer. Moreover, patients find it much easier to take the comparatively small

TABLE 1

The alkaline tide of the urine and associated factors in abnormal cases

Case	Date	Urine																									
		Amount						Specific gravity						Alkalinity													
		Morning						Morning						Night		Morning											
		Night			Total			Night			Morning			11-7		7-8		8-9		9-10		10-11		11-12			
		11-7	7-8	8-9	9-10	10-11	11-12	8-12	11-7	7-8	8-9	9-10	10-11	11-12	11-7	7-8	8-9	9-10	10-11	11-12	11-7	7-8	8-9	9-10	10-11	11-12	
Chronic glomerulonephritis																											
1*	1920	cc	390	33	43	52	45	57	197	cc	1 014					1 012					per cent	37	42	45	48	45	45
	11-3										1 008	1 009	1 010	1 011	1 011	1 012	1 011	1 011	1 009	1 012	per cent	46	64	62	68	63	70
	12-11	950	44	65	92	110	73	340	1 014	1 012	1 013	1 012	1 012	1 012	1 012	1 012	1 012	1 013	1 013	1 013	per cent	30	23	35	23	29	28
	12-9	220	440	60	53	105	91	309	1 010	1 009	1 008	1 008	1 008	1 009	1 009	1 009	1 009	1 009	1 009	1 009	per cent	33	28	31	30	30	33
4*	12-15	465	55	100	95	92	70	357	1 010	1 009	1 008	1 008	1 008	1 008	1 008	1 008	1 009	1 009	1 009	1 009	per cent	33	28	31	30	30	33
	11-16	380	38	45	45	36	32	158	1 012	1 011	1 011	1 013	1 012	1 012	1 012	1 012	1 012	1 012	1 013	1 013	per cent	52	63	69	71	63	67
5*	1921																				per cent	87	88	90	91	95	91
	1-27	325	38	35	44	55	47	181	1 009	1 009	1 008	1 008	1 007	1 007	1 007	1 008	1 007	1 007	1 008	1 008	per cent	87	88	90	91	95	91
6*	5-4	716	48	47	66	77	27	217	1 010	1 012	1 010	1 009	1 010	1 010	1 010	1 009	1 010	1 010	1 010	1 010	per cent	71	59	65	65	72	63
	2-12	216	38	30	30	34	28	122	1 012												per cent	49	52	56		51	50
	2-15	320	53	52	63	53	54	222	1 013	1 012	1 011	1 010	1 010	1 009	1 009	1 010	1 009	1 009	1 010	1 010	per cent	67	73	72	69	72	56
	2-17	255	24	38	48	25	12	123	1 013	1 012	1 009	1 010	1 010	1 009	1 010	1 010	1 010	1 010	1 010	1 010	per cent	45	51	53	50	46	41
7	1920																				per cent	97	89	81	82	68	63
	11-24	325	40	42	128	77	44	291	1 013							1 005	1 006	1 006	1 006	1 006	per cent	97	89	81	82	68	63
	11-26	248	36	45	130	34	35	244	1 017							1 004					per cent	82	73	75	78	85	75
	11-18	190	30	26	65	27	24	142	1 018							1 007					per cent	75	68	70	70	62	64
8	12-7								1 011	1 017	1 004	1 004	1 004	1 004	1 004	1 004	1 004	1 004	1 004	1 004	per cent	73	56	76	80	80	82
	11-18	885	155	125	130	130	113	498	1 007	1 007	1 008	1 007	1 007	1 007	1 007	1 007	1 007	1 007	1 007	1 007	per cent	36	46	46	50	45	48

[illegible]

Maternal hypertension

	17	1-25	290	60	156	148	210	514	1 013	1 009	1 001	1 003	1 005	67	77	85	86	87
18*	2-5	150	90	23	21	10	11	101	1 017	1 017	1 017	1 015	1 015	28	35	43	44	45
19*	2-15	275	22	77	115	29	35	256	1 017	1 017	1 008	1 010	1 015	16	28	32	29	37
20	2-19	68	20	26	38	38	11	78	1 028	1 026	1 026	1 023		24	28	39	16	39

Arteriosclerosis with renal changes

21	1970 11-30	200	35	168	165	90	31	151	1 020	1 005	1 003	1 007		38	60	85	83	83	56
	1971 2-25	285	15	69	115	56	14	281	1 010	1 006	1 003	1 008			32	41	44	35	33
22	1-25	360	19	76	89	08	16	308	1 005	1 006	1 003	1 003		66	81	80	81	77	
23	3-2	505	26	210	215	70	23	518	1 008	1 015	1 005	1 002		63	86	93	88	90	
24	2-1	205	21	61	112	25	37	265	1 020	1 017	1 005	1 012		15	24	41	67	28	22

Essential hypertension

26	19 0	96	32	23	255	25	15	319	1 021	1 015			1 002	1 010	1 011	31	51	36	15	35	13
	19 1																				
27	2-17	80	55	100	108	170	90	168	1 010	1 018	1 007	1 002	1 004	1 002	1 007	18	30	57	75	63	66

TABLE 1—Continued

TABLE 1—Continued

Case	Date	Urine																			Alkalinity			
		Amount								Specific gravity														
		Morning								Night														
		Total								Morning														
		11-7	7-8	8-9	9-10	10-11	11-12	11-7	7-8	8-9	9-10	10-11	11-12	11-7	7-8	8-9	9-10	10-11	11-12					
Nephrosis																								
29	1920 12-17	cc. 280	cc 91	cc 41	cc. 46	cc 50	cc 45	cc. 182	1 025	1 026	1 024	1 021	1 022	1 024	per cent 58	74	90	92	93	91				
Chronic endocarditis with cardiac insufficiency																								
30	1921 3-2	155	24	38	75	74	25	212	1 020	1 018	1 016	1 007	1 007	1 014	36	92	93	93	82	77				
Focal nephritis																								
31	3-11	237	22	32	46	21	23	122	1 019	1 018	1 017	1 015	1 017	1 019	22	22	42	50	39	37				
32	1-25	255	30	65	70	36	34	205	1 015	1 015	1 009	1 007	1 011	1 012	20	30	34	34	32	28				
Gout																								
33	1-26	165	15	95	50	70	40	255	1 019		1 008	1 007	1 009	1 013	27	37	37	33	41	42				
	2-1	135	16	11	7	12	12	42	1 021						30	35	37	32	44	36				
Bilateral polycystic kidneys																								
34	1-28	265	84	33	34	42	46	155	1 013	1 012	1 011	1 011	1 011	1 011	22	23	26	31	25	25				

* Patients who died

TABLE 2
The alkaline tide of the urine and associated factors in normal persons

		Urine																																					
		Amount						Specific gravity						Alkalinity						pH																			
Case	Date	Morning					Total	Morning					Night	Morning					Night	Morning				Night															
		7-8	8-9	9-10	10-11	11-12		7-8	8-9	9-10	10-11	11-12		7-8	8-9	9-10	10-11	11-12		7-8	8-9	9-10	10-11		11-12														
A	1-5	118	21	135	138	125	75	773	1	030	1	029	1	008	1	003	1	010	1	016	21	26	70	85	93	95	5	4	5	6	6	1	6	4	7	3	7	7	
	1-28	187	30	55	304	122	86	567												25	28	87	86	92	93	5	1	5	4	6	8	6	8	6	98	7	25		
B	1-9	164	14	100	240	62	196	598	1	031				1	016	1	011	1	020	1	012	37	35	65	80	94	85	5	7	5	6	6	0	6	0	7	3	6	9
	1-21	210	15	25	62	208	88	383	1	035	1	032	1	032	1	018	1	005	1	012	56	76	76	85	80	94	71	6	2	6	2	7	1	6	8	6	7		
C	1-17	204	76	380	181	102	100	1066	1	021	1	018	1	004	1	003	1	013	1	017	38	40	74	79	94	96	5	7	5	6	6	1	6	2	7	0	7	5	
	1-27	154	30	136	220	110	106	602	1	027	1	030	1	006	1	008	1	010	1	012	23	61	86	91	93	90	5	3	5	4	6	75	6	55	6	8	7	5	
D	1-2	215	21	70	88	60	12	260	1	032				1	018	1	012	1	020	1	022	35	39	18	48	66	64	5	8	5	8	6	0	6	1	6	3	6	2
	1-27	254	62	226	251	71	72	626	1	028	1	032	1	010	1	008	1	020	1	020	26	11	75	76	77	70	5	1	5	7	6	35	6	4	6	75	6	6	
E	1-2	182	31	152	176	102	68	498	1	030	1	030	1	010	1	007	1	016	1	020	41	62	82	93	96	84	5	65	6	05	6	4	6	8	7	3	7	25	

Nephrosis

53	1921	145	16	100	62	18	26	236	1 014	1 005	1 005	1 010	1 010	60	89	91	90	97	92	5 7	6 9	6 7	7 38	6 9
54	1922	150	81	75	70	52	281	1 016	1 010	1 012	1 013	1 011	1 016	36	13	73	65	61	61	5 9	5 95	6 3	6 3	6 1

Malignant hypertension

55	1921	68	17	12	28	23	17	110	1 027	1 025	1 014	1 009	1 011	1 025	30	27	35	30	26	20	5 4	5 3	5 5	5 4	5 2	5 0
56	1-21	136	31	113	72	11	40	299	1 028	1 022	1 008	1 010	1 015	1 021	36	80	88	71	61	61	5 7	6 8	6 9	6 85	6 85	6 7
57	1-26	320	37	38	185	215	62	500	1 015	1 012	1 011	1 005	1 004	1 011	20	41	38	73	63	57	4 8	5 8	5 2	5 9	5 85	5 8
58	1922																									
59	2-8	200	26	92	186	12	34	311	1 018		1 008	1 005	1 012	1 016	36	47	56	55	41	50	5 5	5 75	5 8	6 1	5 85	5 8
59*	3-1	110	135	15	7	80	6	118	1 018	1 016	1 018		1 017		38	66	66	62	62		5 3	6 1	5 95		5 6	

Essential hypertension

60	1921	120	55	72	91	63	31	260	1 010	1 009	1 009	1 004	1 007	1 008	27	40	16	55	41	31	5 1	5 3	5 1	5 8	5 1	5 25
61	5-5	305	30	155	171	50	22	101	1 015	1 015	1 004	1 003	1 009	1 020	21	27	50	67	40	32	5 1	5 3	5 85	5 9	5 6	5 4

Focal nephritis

62	1921	230	30	110	10	29	29	239	1 015	1 013	1 005	1 011	1 015	1 011	39	93	97	86	76	93	5 6	6 8	7 0	6 9	6 7	6 8
63	3-21	155	21	285	200	190	25	700	1 011	1 010	0 031	0 031	0 071	1 011	35	51	85	93	96	98	5 5	5 4	6 7	6 8	7 38	7 7
64	1-16	350	67	35	155	56	65	311	1 011	1 010	1 010	1 007	1 013	1 012	22	67	60	69	19	49	5 0	6 1	6 2	6 7	5 8	5 9
65	1-28	108	18	117	79	76	30	302	1 028		1 007	1 010	1 017	1 019	27	33	83	92	91	85	5 5	5 5	6 8	6 9	6 9	6 9
66	5-7	162		200	307	122	31	663	1 016		1 006	1 004	1 003	1 012	23		90	85	87	82	1 9		6 8	6 1	6 8	6 9
67	5-28	166		16		76		122	1 030		1 021		1 021		33				11		5 1		5 5		5 6	
68	2-27	111	32	11	26	21	18	112	1 026	1 030	1 018	1 022	1 025	1 028	51	61	79	67	11	39	5 5	6 1	6 75	6 3	5 6	5 1
69	3-22	300	38	40	92	56	12	270	1 016		1 013	1 011	1 011	1 017	18	13	16	36	25	26	1 95	5 55	6 5	5 2	5 0	5 0

Case	Date	Amount										Specific gravity				Alkalinity				pH								
		Night					Morning					Night		Morning		Night		Morning										
		Total					Morning					Night		Morning		Night		Morning										
		11-7	7-8	8-9	9-10	10-11	11-12	8-12	11-7	7-8	8-9	9-10	10-11	11-12	11-7	7-8	8-9	9-10	10-11	11-12	11-7	7-8	8-9	9-10	10-11	11-12		
Bilateral hydronephrosis																												
70	1921	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	
	3-31	165	130	65	14	34	52	165	1 007	1 008	1 009	1 008	1 008	91	80	79	75	46	48	7 2	6 85	6 8	5 9	5 8				
Arteriosclerosis																												
71†	1922	2-21	142	98	90	82	106	56	334	1 025	1 024	1 014	1 006	1 007	1 008	24	28	31	38	38	32	5 0	5 0	5 15	5 25	5 1		
Achyia gastrica																												
72	1921	5-13	382	96	330	275	226	77	908	1 014	1 011	1 005	1 005	1 007	1 013	70	98	96			96	6 8	7 6	7 0			7 0	7 1
	5-15	368	78	140	332	98	105	675	1 015	1 015	1 015	1 014	1 005	1 008	1 010	47	88	94	94	95	94	5 9	6 8	6 9	6 85	6 9	7 2	
Polyuria																												
73	3-31	140	15	34	155	185	60	434	1 015	1 012	1 004	1 005	1 011	25	32	69	67	64	76	5 4	5 8	6 5	5 85	5 9	6 7			
Orthostatic albuminuria																												
74	3-25	122	27	110	275	65	33	483	1 021	1 018	1 007	1 003	1 011	1 015	23	46	86	92	98	96	5 2	5 8	6 8	6 8	7 6	7 38		

* Patients who died

† Cases in which the complete study was made

amount of water required in this test. The estimation of the carbon dioxide in the specimens would not seem to be of practical value since it varies in amount directly as the hydrogen-ion concentration.

CONCLUSIONS

1 In this paper are reported the results of investigation of the night specimens and hourly morning specimens of urine in a series of normal and abnormal subjects. At 8 00 a m , 500 cc of water was given on an empty stomach. Most of the abnormal cases were of renal lesions.

2 Normal subjects, as demonstrated by Leathes and confirmed by my experiments, always show a diuresis and an increase in alkalinity during the morning, while abnormal subjects may react like the normal, or show limitation, either partial or complete, of either or both.

3 In addition, among the abnormal subjects examined, there were a few with a high initial alkalinity and an acid tide during the morning.

4 The specific gravity may show little variation even with a diuresis within normal limits, an alkaline tide, or both.

5 Cases with no alkaline tide are usually severe clinically. An acid tide is of grave prognostic import.

6 The curve of the pH figures parallels that of the alkalinity per cent.

7 In the cases reported here, both normal and abnormal, a higher pH figure was always accompanied by increased relative carbon dioxide, and a more acid urine by a decrease in carbon dioxide.

8 The phosphates of normal subjects, as demonstrated by Fiske, have an initial drop followed by a late rise during the morning. In the present series the night specimen was found to have a much lower phosphate content in abnormal than in normal subjects.

9 The results of these investigations show that this test may be of clinical value from the point of view of diagnosis and prognosis.

TABLE 4
Carbon dioxide and phosphorus content of urine in normal and abnormal subjects

[illegible]

Nephrosis

51†	3-	7	27	1	27	1	25	6	18	1	50	2	53	7	80	0	11	1	21	5	36	3	35	1	27	9	65	1	58	1	19	4	21	0	25	3	31	7	190	8	87	2	15	4	15	8	17	7	18	1
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Malignant hypertension

58†	2-	9	1	9	6	7	10	2	19	1	6	7	6	7	9	8	1	7	8	1	35	6	2	8	2	2	97	0	97	0	27	7	15	4	48	0	75	7	194	0	25	2	24	4	28	6	20	2	25	8
59†	3-	1	19	126	6	11	9	5	3	12	2	5	0	85	5	35	9	6	7			9	8	92	9	50	6	45	9	51	5	16	3	62	5	408	9	68	3	20	6		37	0						

Focal nephritis

68†	2-27	12	0	27	5	11	0	33	5	16	7	9	5	16	0	8	8	18	1	8	7	1	0	1	7	125	0	85	5	38	5	51	0	90	9	105	2	142	5	27	1	16	9	13	3	31	8	18	9
69†	3-22	6	8	7	7	9	1	11	2	5	9	5	0	20	4	2	9	7	6	10	3	3	3	2	1	81	6	46	7	23	8	21	3	31	0	52	6	244	9	17	8	19	0	19	6	19	0	22	1

Arteriosclerosis

71†	2-21	13	1	9	1	11	9	12	1	11	2	13	0	18	6	8	3	13	5	9	9	11	9	7	3	104	2	95	1	15	9	16	7	21	7	32	1	147	9	93	5	41	8	13	7	23	0	17	9
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* Patients who died

† Cases in which complete study was made

TABLE 5
Clinical and functional findings in abnormal subjects

Case	Date	Clinical findings						Findings in test					
		Blood pressure	Albumin	Maximal blood urea	Maximal blood creatinin	Edema	Eye-grounds	Phenolsulphone phthalein	Diuresis	Specific gravity range	Alkalinity		
											Initial	Maximal	
Chronic glomerulonephritis													
1*	1920 11- 3	218	1-3	119	5 6	0	Marked retinal edema with many hemorrhages and exudates	5 per cent	197	1 012-1 014	37 per cent	48	
2*	12-11	230	2-3	173	4 9	+	Edema of disc and retina with many hemorrhages and exudates	10	340	1 008-1 011	46	70	
3	12- 9	190	2-3	41	1 7	0	Arteriosclerosis	35	309	1 012-1 014	30	35	
4*	12-15	180	2	231	7 2	0	Normal	5	357	1 008-1 010	33	33	
5*	11-16	196	2-4	139	7 4	+	One hemorrhage, arterial changes	10	158	1 011-1 013	52	71	
	1921 1-27							5	191	1 007-1 009	87	95	
6*	5- 4								197	1 009-1 012	71	72	
	2-12	175	2-3	88 6	2 4	0	Normal	20	122		49	56	
	2-15								222	1 009-1 013	67	72	
7	2-17								123	1 009-1 013	45	53	
	1920 11-24	140	1-3	44	1 58	+	Normal	45	291	1 005-1 013	97	97	
	11-26								244	1 004-1 017	82	85	
8	11-18								142	1 007-1 018	75	75	
	12- 7	158	2-4	30	1 39	+	One small hemorrhage	40		1 004-1 017	73	82	
9	11-18	135	2-3	169	5 9	0	Normal	15	498	1 007-1 008	36	50	

1921	150	2	289	6 2	0	Normal	5	1 007-1 008 1 007-1 009 1 007-1 008 1 008-1 010 1 012-1 013	Acid tide Acid tide Acid tide Acid tide Acid tide
10*	1-4 1-6 1-8 1-12								
11*	1-31	2-3	84	2 48	0	1 dema. of nerve with few hemorrhages	20		36
12	2-7	1-2	128	5	0	Slight arteriosclerosis	10	1 007-1 008	51
13	2-15	0-1	36 2	1 35	Slight	Marked arterial changes	55	1 003-1 020	14
14*	2-18	1-2	75 6	2 59	+	Discs swollen, many hemorrhages and exudates	10	1 007-1 010	59
15	3-4	1-2	51	2 1	0	Normal	30	1 006-1 012	21
16*	3-10	1-2	186	11 2	0	Edema of discs, few exudates	5	1 006-1 007	21

Malignant hypertension

17	1-25	230	1	53	1 54	0	Arterial changes with hemorrhages	45	514	1 003-1 013	67	86
18*	2-5	220	1-2	48	2 0	0	Violent neuroretinitis, nerve heads swollen, hemorrhages and exudates	30	101	1 015-1 017	28	45
19*	2-15	210	1	10	1 7	0	Arterial changes, edema of nerve and retina, few exudates and hemorrhages	45	256	1 005-1 017	16	38
20	2-19	200	0-1	38 7	1 37	0	Marked arterial changes with few exudates and hemorrhages	60	78	1 023-1 028	24	16

Arteriosclerosis with renal changes

	1970	11-40	2-3	35	1-6	0	Old inflammatory changes, marked arteriosclerosis	60	154	1 003-1 020	38	85
21												
22	1971	2-25	150	1	32	1 36	Slight	35	284	1 003-1 011	32	414

TABLE 5—Continued

TABLE J—Continued

Case	Date	Clinical findings						Findings in test				
		Blood pressure	Albumin	Maximal blood urea	Maximal blood creatinin	Edema	Eye-grounds	Phenolsulphone-phthalein	Diuresis	Specific gravity range	Alkalinity	
											Initial	Maximal
Arteriosclerosis with renal changes—Continued												
23	1920 1-25	240	2-3	45.6	2.02	+	Marked arterial changes, edema of nerve and retina, hemorrhages and exudates	45	308	1.003-1.006	66	84
24	3-2	190	1	56.8	1.43	0	Arteriosclerosis with hemorrhages	60	518	1.002-1.015	63	93
25	2-4	165	1	38	1.5	Slight	Normal	50	265	1.005-1.020	15	67
Essential hypertension												
26	1920 1-6	180	0-2	31.2	1.53	Slight	Arteriosclerosis	40	348	1.002-1.021	34	54
27	1921 2-17	150	0	45.9	1.43	0	Normal	60	368	1.002-1.019	18	75
28	3-8	170	0-1	27		0	Moderate arterial changes	65	249	1.011-1.020	39	98
Nephrosis												
29	1920 12-17	130	2-4	36	1.5	+	Normal	40	182	1.021-1.026	58	93
Chronic endocarditis with cardiac insufficiency												
30	1921 3-2	150	1-2	26	1.47	+	Normal	30	212	1.007-1.020	36	93

Focal nephritis											
31	3-11	132	2-3	41 9	1 66	0	Normal	60	122	1 015-1 019	22
32	1-25	110	1-2	32	1 8	0	Normal	72	205	1 007-1 015	20
											34
Gout											
33	1-26 2-1	150	0-1	49	1 86	Slight	Normal	62	255	1 007-1 019	27
									42		30
											44
Bilateral polycystic kidneys											
34	1-28	150	1-2	47 5	1 76	0	Normal	61	155	1 011-1 013	22
											31

* Patients who died

TABLE 6
Clinical and functional findings in abnormal subjects showing fluctuation of alkalinity and hydrogen-ion concentration

Case	Date	Clinical findings						Findings in test							
		Blood pressure	Albumin	Maximal blood urea	Maximal blood creatinin	Edema	Eye-grounds	Phenolsulphone-phthalein	Diuresis	Specific gravity range	Alkalinity		pH		
											Initial	Maximal	Initial	Maximal	
Chronic glomerulonephritis															
35	1921 3-18	210	1-2	50.6	2.68	0	Marked arterial changes with edema and hemorrhages	30	114	1.009-1.017	24	30	5.3	5.2	
36*	4-27	194	1-2	198	8.92	0	Arteriosclerosis	10	194	1.009-1.012	42	52	5.5	5.8	
37	4-27	260	1	47.8	1.77	+	Arterial changes, recent edema, hemorrhages and exudates	30	258	1.008-1.021	22	48	5.4	5.8	
38	4-28	222	3-4	110.5	7.06	+	Fundi anemic	20	287	1.012-1.014	33	44	5.1	5.4	
39	4-29	120	1	33.6	1.36	+	Normal	55	517	1.004-1.018	19	72	5.3	6.8	
40*†	5- 8	192	1-3	74	1.8	+	Normal	40	144	1.011-1.014	81	Acid tide	7.2	Acid tide	
41	5-11	210	1-3	51	1.76	+	Arteriosclerosis	45	111	1.017-1.019	19	33	4.9	5.1	
42*	5-25	170	1-3	101	2.9	Slight	Acute neuroretinitis with edema, exudates and old hemorrhages	30	211	1.009-1.013	35	42	5.0	5.4	
43	5-30	175	1-2	41.5	1.6	+	Pigmentation of retina, slight arteriovenous compression	65	355	1.003-1.023	28	87	5.1	6.9	
44*	5-31	210	1-3	64	1.8	0	Marked retinitis with exudates and hemorrhages	35	157	1.013-1.017	26	42	5.0	5.3	
45*	6-30	242	0-2	56	1.9	+	Intense neuroretinitis with exudates and hemorrhage	40	113	1.006-1.015	72	81			

[illegible]

Subacute diffuse nephritis

[illegible]

Nephrologis

53	3-12	139	2-4	26 8	1 5	+	Normal	40	236	1 005-1 011	60	97	5 7	7 38
54	10 ¹²													
54	3-7	110	1	37	1 5	0	Normal	50	281	1 010-1 016	36	72	5 9	6 4

Malignant hypertension

55	1971	220	1	33	8	1	25	0	Slight arterial changes	15	110	1 009-1	027	30	35	5 4	5 5
56*	1-21	260	2	16	1	14	0	0	Severe neuroretinitis with hemorrhages and exudates	50	299	1 008-1	028	36	98	5 7	6 9
57*	1-26	210	1	67	1	97	0	0	Arterial changes with retinitis, exudates and hemorrhages	35	500	1 004-1	015	20	73	4 8	5 9
58†	2-4	235	1-3	51	1	6	0	0	Arterial changes, slight edema, exudates	15	311	1 005-1	018	36	56	5 5	6 1
59*†	3-1	234	1-3	30			0	0	Arterial changes, edema, exudates and hemorrhage	30	138	1 016-1	015	39	66	5 3	6 1

Arteriosclerosis

	1922	108	0-1	26		0	Normal	50	334	1 006-1 025	24	38	5 0	5 25
71	2-21													

Achyia gastrica

	1921													
72	5-13	171	0	11	1 3	0	Normal	50	908	1 005-1 014	70	96	6 8	7 6
72	5-15	171	0	11	1 3	0	Normal	50	575	1 005-1 015	17	95	5 9	7 2

Polyuria

	1921	130	0-1	25	1 1	1 39	0	Normal	65	131	1 001-1 015	25	76	5 1	5 9
73	3-31														

Orthostatic albuminuria

	1925	120	0-2	15	1 45	0	Normal	60	483	1 003-1 021	23	98	5 2	7 6
71	3-25													

* Patients who died

† Cases in which complete study was made

TABLE 7

Carbon dioxide combining power of blood plasma

Case	Diagnosis	Before diuresis	After diuresis
A	Normal	58.9	56.0
B	Normal	59.8	55.1
C	Normal	62.6	59.8
D	Normal	60.7	60.7
E	Normal	67.3	67.3
F	Normal	67.3	67.3
G	Normal	59.6	59.6
46	Normal	71.5	70.6
46	Chronic glomerulonephritis	55.9	55.9
47	Chronic glomerulonephritis	61.4	63.3
48	Chronic glomerulonephritis	50.8	50.8
54	Nephrosis	66.0	67.8
58	Malignant hypertension	62.6	62.6
59	Malignant hypertension	77.0	74.0
68	Focal nephritis	67.3	67.3
69	Focal nephritis	71.6	67.8
71	Arteriosclerosis	69.6	69.6

TABLE 8
Findings in cases with an acid tide*

Case	Urine																								
	Amount						Specific gravity						Alkalinity						pH						
	Morning						Night						Morning						Night						
	11-7	7-8	8-9	9-10	10-11	11-12	11-7	7-8	8-9	9-10	10-11	11-12	11-7	7-8	8-9	9-10	10-11	11-12	11-7	7-8	8-9	9-10	10-11	11-12	
10	Chronic glomerulonephritis																								
	1921	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	cc	
	1-1	160	92	95	65	80	65	1 005	1 000	1 007	1 008	1 001	007	98	97	94	85	71	71						
	1-6	215	100	75	60	60	70	1 009	1 009	1 008	1 008	1 007	1 008	93	95	73	62	52	52						
	1-8	345	61	55	60	75	80	1 008	1 008	1 007	1 007	1 007	1 007	98	98	92	69	58	58						
70	Bilateral hydronephrosis																								
	1-12	340	11	47	10	31	37	1 009	1 009	1 008	1 009	1 009	1 010	60	14	11	44	40	39						
	3-31	165	130	65	11	31	52	1 007	1 008	1 009	1 008	1 008	91	81	79	75	47	19	19						
	7-8	165	130	65	11	31	52	1 007	1 008	1 009	1 008	1 008	91	81	79	75	47	19	19						
	11-12	165	130	65	11	31	52	1 007	1 008	1 009	1 008	1 008	91	81	79	75	47	19	19						
10	Chronic glomerulonephritis																								
	5-8	292	10	18	18	32	16	1 011	1 011	1 011	1 011	1 013		91	61	53	19	19	57	7 2	5 9	5 5	5 4	5 35	5 5
	10-2	292	10	18	18	32	16	1 011	1 011	1 011	1 011	1 013		91	61	53	19	19	57	7 2	5 9	5 5	5 4	5 35	5 5
	2-3	251	35	35	10	32	34	1 016		1 015	1 011	1 015	69	71	69	65	67	63	63	6 9	6 6	6 1	6 1	6 1	6 0
	10-2	251	35	35	10	32	34	1 016		1 015	1 011	1 015	69	71	69	65	67	63	63	6 9	6 6	6 1	6 1	6 1	6 0
16	Chronic glomerulonephritis																								
	2-3	251	35	35	10	32	34	1 016		1 015	1 011	1 015	69	71	69	65	67	63	63	6 9	6 6	6 1	6 1	6 1	6 0
	2-3	251	35	35	10	32	34	1 016		1 015	1 011	1 015	69	71	69	65	67	63	63	6 9	6 6	6 1	6 1	6 1	6 0
	2-3	251	35	35	10	32	34	1 016		1 015	1 011	1 015	69	71	69	65	67	63	63	6 9	6 6	6 1	6 1	6 1	6 0
	2-3	251	35	35	10	32	34	1 016		1 015	1 011	1 015	69	71	69	65	67	63	63	6 9	6 6	6 1	6 1	6 1	6 0

• **Practicality**

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ORTHOPNOEA AND THE EFFECT OF POSTURE UPON THE RATE OF BLOOD FLOW

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(Received for publication July 11, 1925)

Lindhard (1) has noted that, of seven subjects three, all of them females, had a slower rate of blood flow in the sitting than in the recumbent posture while the four male subjects showed practically no change. As far as we know these are the only direct observations in the literature concerning the effect of posture on blood flow.

We have made thirteen determinations of the circulatory rate with the subjects sitting and nine determinations with the subjects standing, each one compared with a preceding determination made while the subject was recumbent. A rest period of about an hour preceded the first determination in each experiment. Change of position was accompanied by as little exertion as possible and a further rest period of fifteen to twenty minutes preceded each determination in the new position. The method used is a gasometric one, based on the Fick principle, which we have previously described (2). The subjects were young, healthy, adult males except the last two who were females and G. R. who has a rheumatic cardiac lesion which is well compensated. None of the subjects were athletic.

Our results, shown in table 1 are even more striking than those of Lindhard.

In no case did we fail to observe a diminution in the rate of blood flow on changing from the recumbent to the sitting position. Only once was the reduction less than 500 cc per minute, while another observation on the same individual showed a greater change.

The slowing of the circulation was more marked in the standing than in the sitting position. The average rate of blood flow while sitting was 76 per cent and while standing it was 50 per cent of the

¹ Aided by the Tutorial Fund of the Harvard Medical School, Boston, Mass.

rate while reclining Inasmuch as the pulse rate increased from an average of 63 per minute in the reclining position to 65 per minute in the sitting position, and 90 per minute in the standing position, the output per beat was tremendously diminished in the upright posture While standing it amounted to only 35 per cent (on the average) of

TABLE 1

Subject	Lying			Sitting*			Standing		
	Pulse	Circulation per minute	Output per beat	Pulse	Circulation per minute	Output per beat	Pulse	Circulation per minute	Output per beat
	<i>per minute</i>	<i>liters</i>	<i>cc</i>	<i>per minute</i>	<i>liters</i>	<i>cc</i>	<i>per minute</i>	<i>liters</i>	<i>cc.</i>
A V B	66	5 11	77				88	3 95	45
	65	6 49	100	68	5 73	84			
	68	6 30	92 5	68	5 80	85	78	4 46	57
H F	73	9 87	136				95	4 67	49
	72	9 60	133	80	7 29	91			
	68	8 66	131	70	8 42	120	89	3 90	44
A C R	60	8 90	148	64	3 75	59	83	4 12	50
S A O	54	9 73	180	66	7 08	107	79	4 42	56
	58	6 52	112	59	5 81	98 5			
J R L	56	9 04	161	54	4 92	91	76	3 12	41
W A Mc	55	8 12	148	57	5 66	99			
K M M	58	6 69	115	58	5 28	91	105	3 41	32 5
G R	62	10 90	176	56	8 07	144			
L G	62	8 27	133	66	7 40	72			
M E M	62	6 42	103 5	80	4 21	53 5	115	4 05	35
Average	63	8 04	127 6	65	6 11	94 0	90	4 01	44 6
Per cent of reclining value				103	76	74	143	50	35

* The values obtained for the blood-flow in the sitting position are not unlike those published by Burwell and Robinson in volume one of this Journal Their determinations were made upon subjects at rest in a reclining position in a hospital wheel chair

that while reclining This is also indicated by the blood pressure findings After the subject had been standing for a time the pulse pressures were greatly diminished, often to 5 or 10 mm of mercury, and sometimes the auscultatory vascular sounds were almost inaudible

As a part of a study of circulatory and respiratory conditions in

college girls, many of whom were especially athletic, Miss Abby H Turner (3) has made similar determinations in the laboratory of Physiology of the Harvard School of Public Health, using our method. Of 32 determinations of blood flow in the sitting position, 8 showed a rise, 7 little or no change and 17 a distinct fall from the resting volume. Of 19 determinations made in the standing position, 1 showed a larger volume of blood flow than in the reclining position, 4 showed practically no change, and 14 a smaller volume flow.

Y Henderson and Haggard (4) have recently repeated these experiments by a new method and have obtained quite similar results. As the phenomenon has thus been observed in four different laboratories by three different methods, there can hardly be any question as to its occurrence. Its cause and its significance, on the other hand, are interesting subjects for speculation.

DISCUSSION

The obvious explanation is to attribute the change in blood flow to an effect of gravity which, in the upright position, produces a stasis of the circulation in the legs and splanchnic area which results, consequently, in a diminished supply of blood to the heart. It is well known that holding a rabbit suspended by the ears will in a short time result in syncope and, after a variable time, in death. Soldiers sometimes faint during long periods of standing at attention. We have noted that while a subject was standing during an experiment the calves of his legs became indurated and brawny and increased in circumference about 1.5 cms.

L Hill (5) and Hill and Barnard (6) have studied the effect of posture on the arterial and venous blood pressures of animals. They found that the normal dog was able to compensate, in part, for the effect of gravity and to maintain a circulation in the vertical position with feet down although with a considerable drop in the blood pressure of the arterial system and of the superior vena cava. When the splanchnic vaso-motor control and the respirations were inhibited, however, assumption of the vertical feet down position was followed by a drop in the arterial blood pressure to zero and, in a short time, in death of the animal.

Hill found that the vertical position had much less effect on the blood pressure in monkeys than in dogs, and concluded that their compensatory mechanisms were more highly developed. Our experiments might be taken to indicate that even man is not completely adjusted to the upright position. Or perhaps one of the adjustments consists in a rate of blood flow which, in the reclining position, is so much more rapid than the needs for gas transport demand, that the slower flow in the upright position is still within the limits of safety so that no difficulty usually develops.

The diminution of the arterial pulse pressure in the upright position has been noted. We have made a considerable number of determinations of the blood pressures in arm and hand veins with the subjects in the different positions. Both the indirect method of Hooker (7) and a direct method with a needle inserted into a vein and attached to a manometer have been used. The results have been indeterminate. No great changes in venous blood pressures have been found associated with change in position. Nor has the direction of variation been constant. More often there has been some rise in the venous blood pressure after assuming the upright position. Barach and Marks (8) have reported similar changes in arterial and venous blood pressures associated with change in position.

A review of previously reported studies, however, indicates that this need be neither very surprising nor disturbing. Kroetz (9) after a long series of determinations in patients came to the conclusion that there were too many unknown factors influencing the venous blood pressure to attach much significance to it in relation to the general circulation. Wiggers (10) found that, although the return flow of blood was reduced in the early stages of shock, the venous blood pressure remained unchanged or increased and has stated (11) that "as long as the return flow of blood is not reduced too greatly the mean auricular pressure may be maintained by compensatory mechanisms." The association of high venous blood pressures with diminished rates of blood flow, which we have observed to occur in the upright position in man, is a demonstration of such compensatory mechanisms. There is a certain amount of evidence of venomotor activity. The subject needs further investigation. Cardiac tonus might also be a factor

in maintaining the venous blood pressure but the existence of such a tonus has not been proved

The observations herein reported have led us to a consideration of the present conceptions concerning orthopnoea and the possible relationship between the two phenomena—slower blood flow and more comfortable breathing in the upright position

L Hill (5) in 1895 expressed an idea which for a time we thought was original with us. He said "The position selected by patients suffering from lung or heart disease can be explained, for it is manifest that the upright position will afford the greatest relief by diminishing pulmonary congestion through the retention of a large quantity of blood in the splanchnic area." Since that time there have been numerous studies of dyspnoea and orthopnoea yet the statement has frequently been made that there is no satisfactory explanation for orthopnoea. We have seen no further reference to Hill's suggestion.

Concerning the details of the production of dyspnoea itself there may still be some debate. The resting metabolism is frequently found to be increased in dyspnoeic cardiac patients as shown by Peabody, Meyer and DuBois (12), and by Peabody, Wentworth and Barker (13). This increases the demand for respiratory exchange. The respiratory exchange is carried on less efficiently in such patients than in normal persons. They need to breathe a larger volume of air per minute in order to accomplish the same carbon-dioxide elimination and oxygen absorption. Siebeck (14) argued that this was due to an inadequate mixture of air in the lungs and an increase in the functional dead space. Peters and Barr (15) thought that there is an increased tension difference between the gases in the alveolar air and the arterial blood because of a diminished diffusion capacity of the lungs. Peabody, Sturgis Barker and Read (16) unlike Siebeck and Peters and Barr, found that in their cases, although the total ventilation was considerably increased, because of the fact that breathing was more rapid and shallow than normal, the effective ventilation (the total ventilation minus the total dead space air) was not increased out of proportion to the metabolism. Consequently they did not need to invoke either of the above mentioned mechanisms. Their subjects were not as dyspnoeic as those of the

previous workers and it may well be that inefficient gas exchange in the lungs is a late development

Finally the ability of the dyspnoeic cardiac patient to increase the volume of his ventilation is diminished. It has been known for a long time that the vital capacity of cardiac patients is reduced. Peabody and Wentworth (17) demonstrated that the amount of the reduction is closely related to the degree of dyspnoea that the patient suffers. Sturgis, Peabody, Hall, and Fremont-Smith (18) have given the relationship a more exact expression by showing that the maximum ventilation both in normal subjects and in cardiac patients is obtained by increasing the respiratory rate to about thirty-five per minute and the depth of respiration to about one-third of the vital capacity.

Behind all of these factors, however, it seems quite certain, although perhaps not absolutely proven, that the fundamental cause of dyspnoea is congestion of blood in the pulmonary circuit. Basch in 1891 (19) proposed the idea that the increased turgidity of the congested pulmonary vessels diminished the flexibility of the lungs. This is supported by the researches of Lundsgaard (20) who found that the earliest change in the lung volumes of cardiac patients was an increase in the residual air. This occurred before there was any change in the total capacity and was therefore the cause of the early decrease in the vital capacity of the lungs. Peabody, Sturgis, Barker, and Read (16) found that compensated cardiac patients, with little reduction in their vital capacities, had increased respiratory volumes per unit of metabolism because of more rapid, shallow breathing, with a consequent increase in the proportion of dead space air to the total expired air. This they suggest is possibly explained by an earlier excitation of the Hering-Breuer reflex due to the increased rigidity of the lungs.

Greater degrees of pulmonary congestion cause encroachment upon the air space in the lungs. Romanoff (21) found that following increases of pressure in the pulmonary circuit the lungs expanded if the pleurae were opened, but that if such expansion were restricted by intact pleurae the alveolar air space was contracted. Drinker, Peabody and Blumgart (22) and Drinker and Agassiz (23), found that following compression of the pulmonary veins, in their preparation with intact pleurae, there was an increased blood pressure in the

pulmonary artery, while a diminished proportion of air from a constant delivery respiration pump entered the lungs and an increased proportion passed through an overflow tube. Such observations are thought to have a direct bearing on the diminished vital capacities of cardiac patients.

Bohr in 1907 (24) found that the vital capacity is less in the reclining position than it is while standing. He reported only a small series of observations, and was chiefly interested in changes in the mid-capacity. Christie and Beams (25) have recently determined that the average vital capacity of 290 normal people was 5.5 per cent less while lying than while sitting. They also found (26) that in 19 patients with diminished vital capacities but without orthopnoea this difference was 4.7 per cent, in 14 patients with "orthopnoea of choice" it was 12.5 per cent, while in 9 patients with "orthopnoea of necessity" it was 26.7 per cent. There was not only this very great increase in the relative difference between the vital capacities, while lying and sitting in the latter group, but also an increase in the absolute difference of about 50 per cent. Christie and Beams believe that this effect of posture on vital capacity is the primary factor in the production of orthopnoea.

It remains to consider the fundamental cause that is responsible for this effect. For years the chief explanation for orthopnoea has been a lowering of the diaphragm and a supposed increase in the capacity of the chest in the sitting posture. With the descent of the ribs and the narrowing of the intercostal angle that occurs it is questionable if the capacity of the chest is not actually diminished in this position. At any rate, even if such an anatomical relationship were responsible for part of the difference between the vital capacities, lying and sitting, it would not explain the increase in this difference in patients suffering from heart disease. There must be some other factor which is peculiar to heart disease and which is susceptible to the influence of change of position.

It has been argued that the sitting posture permits better use of the muscles of respiration. Such an explanation has never been accepted as satisfactory. Moreover, Peabody and Sturgis (27) have shown that even extreme general weakness does not greatly diminish the vital capacity. Also frequently repeated determinations of the vital ca-

capacity in cardiac patients, a procedure likely to cause considerable fatigue, does not result in its further reduction

It seems much more reasonable to regard as important the condition which is primarily responsible for the reduced capacity of the lungs, that is, the pulmonary circulation. Many researches, too numerous to analyze here, have shown the great susceptibility of the pulmonary blood pressure to various influences. Plumier (28) demonstrated that compression of the inferior vena cava causes a reduction in pulmonary arterial pressure. Pressure on the abdomen by increasing the blood supply to the heart raises it. Cloetta and Staubli (29) found that compression of the aorta or a quick infusion of 15 cc of salt solution into a cat, not only caused a rise in pulmonary arterial pressure but an increase in the volume of the lung in a plethysmograph, which if the lung were encased in a non-expansive box, would greatly diminish the alveolar air space. Fuhner and Starling (30) found that increased venous supply and increased peripheral resistance raised the pulmonary arterial pressure in their heart-lung preparations. Drinker and Agassiz (23) produced similar rises by compression of the pulmonary veins.

The diminished output of the heart which we have demonstrated occurs in the upright position is admirably adapted to reduce the congestion in the lungs, to increase the vital capacity and to make breathing more comfortable. It should give relief in much the same way as does venesection. Instead of the total volume of the blood being reduced in volume, a part of it stagnates in the dependent parts, a smaller amount is returned to the heart, the output of the right heart is diminished, this smaller amount of blood should be more readily dealt with by the left heart and pumped on into the systemic circulation, and the congestion in the lungs should be relieved. A degree of cardiac rest also results from the reduced output of blood.

It will undoubtedly appear strange at first sight that a dyspneic cardiac patient, with a circulation which is already slower than normal, should be relieved by still further slowing of the circulation. It may be pointed out that the normal resting blood flow in man is much faster than the needs for gas transport demand so that there is a wide margin of safety. The tissues, aside from those of the nervous system, stand the anoxemia and the relatively slight increase in

carbon-dioxide tension, which would result from a slow circulation, comparatively well. The nervous system, on the other hand, has such a large blood supply in proportion to its metabolism that its blood is never subject to very great venous change. This being so a slower blood flow does not cause an increase in the respiratory stimulus as long as the blood does not leave the lungs less well ventilated. The respiratory center has such a large blood supply that the capillary blood which controls it is practically arterial blood. Slower blood flow should not therefore impair the ventilation of the arterial blood. A small volume of blood that has been highly utilized, can be as readily freed of carbon dioxide, and as readily oxygenated by the respiratory mechanism as a larger volume of blood that has been less completely utilized. The respiratory demand is a function of the metabolism rather than of the circulation.

Clinical confirmation of this fact is frequently observed. Fainting rather than dyspnea is the reaction to a diminished blood flow. Mediastinal lesions, that do not involve the pulmonary circulation, may cause marked venous engorgement, cyanosis and stasis of the circulation in the upper part of the body without being necessarily accompanied by any considerable degree of dyspnea.

Throughout this paper the argument has been presented from the standpoint of the cardiac patient. The discussion also applies to persons who have dyspnea and orthopnea because of diminution of the vital capacity due to other causes than heart disease. As Christie and Beams (26) pointed out even the 55 per cent increase in vital capacity which normal subjects gained by changing from the lying to the sitting position may mean the difference between extreme embarrassment of the respiration and comparative comfort. It is also quite possible that lesions other than those of the heart, which reduce vital capacity, may also cause pulmonary congestion that may be relieved in the same manner.

SUMMARY

Data showing a diminution in the rate of blood flow in the upright position, thought to be due to an effect of gravity, impeding the return of venous blood to the heart have been presented.

The manner in which the diminished blood flow in the upright

position may reduce dyspnea by relieving the congestion in the pulmonary circulation, has been discussed

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THE ELIMINATION OF CARBON MONOXIDE FROM THE BLOOD

A THEORETICAL AND EXPERIMENTAL STUDY

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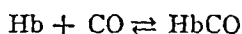
(Received for publication July 11, 1925)

The symptoms attending the partial or complete saturation of the hemoglobin of the blood with carbon monoxide are too well known to require detailed comment. With as little as 40 per cent of the hemoglobin combined with carbon monoxide there may ensue deep coma, marked central nervous system symptoms with ankle clonus, positive Babinski and Kernig signs and often death. Occasionally the blood becomes free of carbon monoxide, the coma terminates, but the patient subsequently sinks into coma again and dies, probably as a result of central nervous system damage.

The exact explanation of this train of symptoms is not entirely clear. Presumably the effects of carbon monoxide asphyxia are those of anoxemia. For, as is well known, anoxemia long continued results in irremedial damage to nervous tissue. This anoxemia is not due primarily to a reduction of the total oxygen of the blood as is commonly supposed, but rather to a marked lowering of the partial pressure at which oxygen is available for tissue metabolism. As will be shown subsequently this effect is the result of a marked alteration in the form of oxygen dissociation curve of blood by carbon monoxide. In the light of this alteration the desperate condition of a subject whose blood is half saturated with CO as contrasted with the comparatively comfortable condition of one who as in anemia cases, has merely lost half his hemoglobin is understandable.

That the effects of carbon monoxide are entirely those of anoxemia is at present an assumption based upon the fact that its toxic effects are supposedly nil. This will be discussed below.

Carbon monoxide, as contrasted with oxygen, has a peculiarly great affinity for hemoglobin. A very small concentration of carbon monoxide in the alveolar air will keep the blood highly saturated with CO. CO at a few millimeters of partial pressure has the same combining capacity as oxygen at a partial pressure of several hundred millimeters. This peculiarity of CO is as yet unexplained, but it enables a small concentration of carbon monoxide in the alveolar air to keep the blood highly saturated with CO and so lead to dangerous asphyxiation. Conversely the recovery of asphyxiated subjects is slow. The elimination of CO from the blood depends upon the reversal of the reaction



Under the ordinary circumstances of recovery the conditions favor the establishment of an equilibrium toward the right. The blood gives up a small amount of CO to the alveolar air. But this small amount is sufficient to reestablish equilibrium at a point close to the original saturation. Part of the CO is removed from the alveolar air at each respiration it is true but as a rule the respirations are depressed. A kind of vicious cycle is set up. The two factors, great affinity of CO for hemoglobin and subnormal respiration, lengthen the time of recovery to hours. Unless the CO be rapidly eliminated in a subject with over 60 per cent of saturation, death usually occurs.¹

The inhalation of CO₂ with pure oxygen effects this rapid elimination. First used by Henderson (1921) its use in many hospitals and particularly in mine rescue stations has demonstrated its value.

Henderson and subsequently Yant and Sayre (1922) showed in experimental animals and subjects that the addition of CO₂ to the air or oxygen inhaled shortened the time of elimination of CO frequently to as brief a time as one-half hour. But these investigators attributed the action of CO₂ to the increased pulmonary ventilation which it

¹ It has been erroneously supposed by some that the velocity of reaction of CO with Hb is a factor, i.e., reaction (1) goes with great speed toward the right and slowly toward the left. Hartridge (1922) has shown that the reaction is practically instantaneous in either direction. If the conditions are properly selected the reaction in vivo can be made to go with great rapidity in either direction as the subsequent protocols will show.

caused This increased ventilation washed out the CO from the alveolar air rapidly, hastening the elimination from the blood by lowering the alveolar CO pressure to a minimum But increased ventilation may not be the sole or even the important factor operating when CO₂ was inhaled by asphyxiated animals The increased hydrogen ion concentration of the blood during CO₂ inhalation must be taken into account and this was not considered by either Henderson or Yant and Sayre

Theoretical considerations of the equilibrium relations of oxyhemoglobin reduced hemoglobin, carbon monoxide hemoglobin, oxygen carbon monoxide and hydrogen ion concentration, supported by experimental evidence, have enabled us to predict that an increase of the acidity of the blood by any agent, ventilatory rate remaining constant, would result in an increased rate of elimination of CO from the blood Experimental rates of elimination under these conditions were found to be quite in accord with the prediction

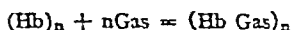
In addition, the relations developed showed that the primary effect of carbon monoxide was a profound alteration of the oxygen dissociation curve, which, rather than the mere loss of functioning hemoglobin, was the cause of the anoxemia

EQUILIBRIUM BETWEEN Hb, HbO₂, HbCO, O₂, CO AND H⁺ THEORY

Two assumptions are made to develop an expression for the effect of CO on the combination of oxygen with hemoglobin

1 Hemoglobin combines with a mixture of O₂ and CO as if they were one substance due regard being paid to the relative affinity of the two gases I e, a partial pressure P_{CO} of CO is equivalent to a partial pressure (K'_{CO/O} P_{CO}) of oxygen, K'_{CO/O} being a constant unaltered for a given blood by any condition save temperature It expresses the relative affinity or saturating capacity of CO for hemoglobin as compared to oxygen At 38°C in human blood it has a value of 300 ÷ 50 as experimentally determined by Barcroft, Douglass and many others In other words, a gas mixture containing P_{O₂} mm of O₂ and P_{CO} mm of CO would be equivalent to [Gas] = (P_{O₂} + K'_{CO/O} P_{CO}) mm. of O₂ in its combining value toward hemoglobin

Hence for the reaction



we have by mass action

$$\frac{[\text{Hb Gas}]_n}{[1 - [\text{Hb}]_n] [P_{O_2} + K'_{CO/O} P_{CO}]^n} = K_e \quad (1)$$

Here [HbGas] represents the fraction of total hemoglobin combined with O_2 and CO

2 The ratio of HbO_2 to $HbCO$ in any mixture of hemoglobin is proportional to the partial pressure of the two gases in equilibrium with it i e ,

$$\frac{[HbO_2]}{[HbCO]} = \frac{P_{O_2}}{K_{co/o} P_{co}} \quad (2)$$

These assumptions were likewise employed by Hill (1921) in the development of an hypothesis which postulated the dissociation of hemoglobin into a non-protein fraction combining with oxygen or carbon monoxide and a protein fraction not capable of combination with either

Equation 2 is known to be true for the case where the hemoglobin is completely saturated with gas But it is as yet undetermined whether this relation holds true when the hemoglobin is *not* completely saturated, i e , when there is still some reduced hemoglobin present However, the validity of this relation for partially saturated hemoglobin is here assumed The identity of $K_{CO/O}$ and $K'_{CO/O}$ remains to be proved

Since $[Gas] = P_{O_2} + K'_{co/o} P_{co}$ and $[HbGas] = [HbO_2] + [HbCO]$, we have

$$K_g = \frac{[HbO_2] + [HbCO]}{[1 - ([HbO_2] + [HbCO])] [P_{O_2} + K'_{co/o} P_{co}]^n} \quad (3)$$

If $P_{co} = 0$

$$\begin{aligned} K_g &= \frac{[HbO_2]}{(1 - [HbO_2]) P_{O_2}^n} \\ &= K_o \end{aligned}$$

i e , Hill's oxygen dissociation constant

Further if $P_{co} = 0$

$$K_g = K_o = \frac{[HbCO]}{K'_{co/o} (1 - [HbCO]) P_{co}^n}$$

but since

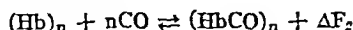
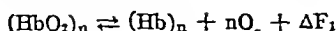
$$K_{co} = \frac{[HbCO]}{(1 - [HbCO]) P_{co}^n}$$

we have by division

$$\frac{K_{co}}{K_o} = K_{co/o}^n \quad (4)$$

A similar relation involving $K_{co/o}$ may be derived thermodynamically and independently of any considerations of the kinetics of the reactions concerned

Consider the following equilibrium states



where ΔF_1 , ΔF_2 and $n \Delta F_3$ are the free energy changes of the respective reactions
From the known equilibrium constants we have

$$-\log K_o = \Delta F_1$$

$$\log K_{co} = \Delta F_2$$

$$n \log K_{co/o} = n \Delta F_3$$

Since $\Delta F_1 + \Delta F_2 = n \Delta F_3$, we have

$$-\log K_o + \log K_{co} = n \log K_{co/o}$$

or

$$\frac{K_{co}}{K_o} = K_{co/o}^n \quad (4')$$

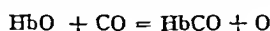
It follows from 4 and 4' that $K'_{co/o} = K_{co/o}$

Experimental proof of (4) is offered by the data on the blood of C A Douglass (1912) Figures 1, 3 and 4 of their paper give the following data n is taken as 2.5

Hemoglobin constants of Douglass blood at 40 mm CO_2 and 38°C

K_{co}	Obs	25000
K_o	Obs	0.031
$K_{co/o}$	Obs	250
$K_{co/o}$ Calculated by equation 4'		230

The experimental and theoretical proof of the identity of the constant $K'_{co/o}$ by which the partial pressure of CO is multiplied to give its combining capacity in oxygen terms with the equilibrium constant $K_{co/o}$ of the reaction



of which equation 2 is the mass action expression permits the introduction of equation 2 into (3)

Eliminating P_{co} we obtain, remembering that $K_g = K_o$

$$K_o = \frac{[\text{HbO}]^n}{([\text{HbO}] + [\text{HbCO}])^{n-1} (1 - ([\text{HbO}] + [\text{HbCO}])) P_{\infty}^n} \quad (5)$$

This equation was tested experimentally on the blood of W C S. The blood was equilibrated at 38° with amounts of CO and O_2 necessary to give partial

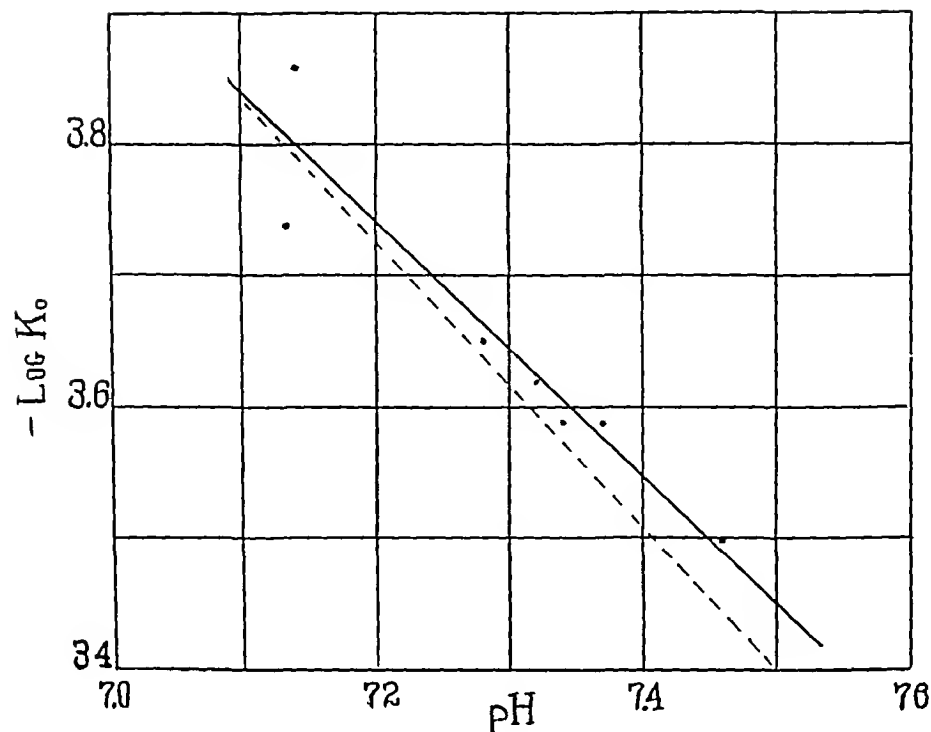


FIG 1 SOLID LINE = LOG K₀ AT VARYING pH CALCULATED BY EQUATION 5 IN BLOOD PARTIALLY SATURATED WITH CO AND O₂
DASHED LINE = Log K₀ determined at varying pH in the presence of O₂ only

TABLE 1

Date	Oxygen Tension	HbO	Satn O ₂	HbCO	Satn CO	pH calculated	-log K ₀ calculated by equation 5
1922-23	mm	volumes per cent	per cent	volumes per cent	per cent		
December 7	20 3	7 3	36 5	6 9	35 0	7 28	3 65
December 13	18 6	6 9	32 0	8 3	38 6	7 32	3 62
February 15	11 7	4 6	21 8	6 8	32 4	7 34	3 59
April 7	20 9	8 1	38 5	8 8	42 0	7 37	3 59
April 7	20 9	6 3	30 0	8 1	38 5	7 15	3 86
April 11	23 3	8 3	41 3	8 1	40 5	7 46	3 50
	19 3	6 1	30 3	6 5	32 5	7 15	3 73
June 6	6 7		25 5		57 0	7 34	

saturation of the hemoglobin at varying pH values The per cent saturation with O₂ and CO, the oxygen tension and pH were determined using the Van Slyke-Stadie, Haldane and Cullen method respectively The values for K₀ thus obtained

are compared with K_o determinations using oxygen alone² The results are given in figure 1 and table 1 There is excellent agreement.

EFFECT OF CHANGE OF pH OF THE BLOOD ON THE OXYGEN DISSOCIATION CURVE IN CARBON MONOXIDE POISONING

By means of Equation (5) the oxygen dissociation curve of whole blood may be drawn when the blood is *constantly* saturated with CO to any definite and fixed degree Fixing the CO saturation at 50 per cent we have for an average human blood the following normal conditions as contrasted with an acidotic condition easily attained by inhalation of CO_2

	I	II
pH	7.4	7.0
K_o	0.001	0.0009
$K_{co/o}$	250	250
n	2.2	2.2
HbCO	50 per cent	50 per cent

In figure 2 are shown the curves for these two conditions calculated from equation 5

Increasing the acidity of the blood changes K_o only according to the well established (over physiological ranges) relation

$$\Delta \log K_o = \Delta pH$$

$K_{co/o}$ and n are unaltered as has been shown by Barcroft and others

Figure 2 shows that the presence of CO has changed markedly the form of the oxygen dissociation curve of that part of the hemoglobin uncombined with CO

The effect of this alteration upon the rate of elimination of CO from the blood as influenced by increasing the hydrogen ion concentration only is clearly seen from the following

At point A we have from figure 2

pH	= 7.4
HbCO	= 50 per cent
HbO ₂	= 40 per cent
P_{co}	= 0.147 mm Hg
P_{o_2}	= 30 mm Hg

² The control K_o values determined with oxygen alone are taken from published data Stadie and Martin, Jour Biol Chem, 1924, lx, 191

Lower the pH to 7.0 and maintain the oxygen tension constant
Then at point B we have

$$\begin{aligned} \text{pH} &= 7.0 \\ \text{HbCO} &= 50 \text{ per cent} \\ \text{HbO}_2 &= 32 \text{ per cent} \\ P_{\text{CO}} &= 0.167 \text{ mm Hg} \\ P_{\text{O}_2} &= 30 \text{ mm Hg} \end{aligned}$$

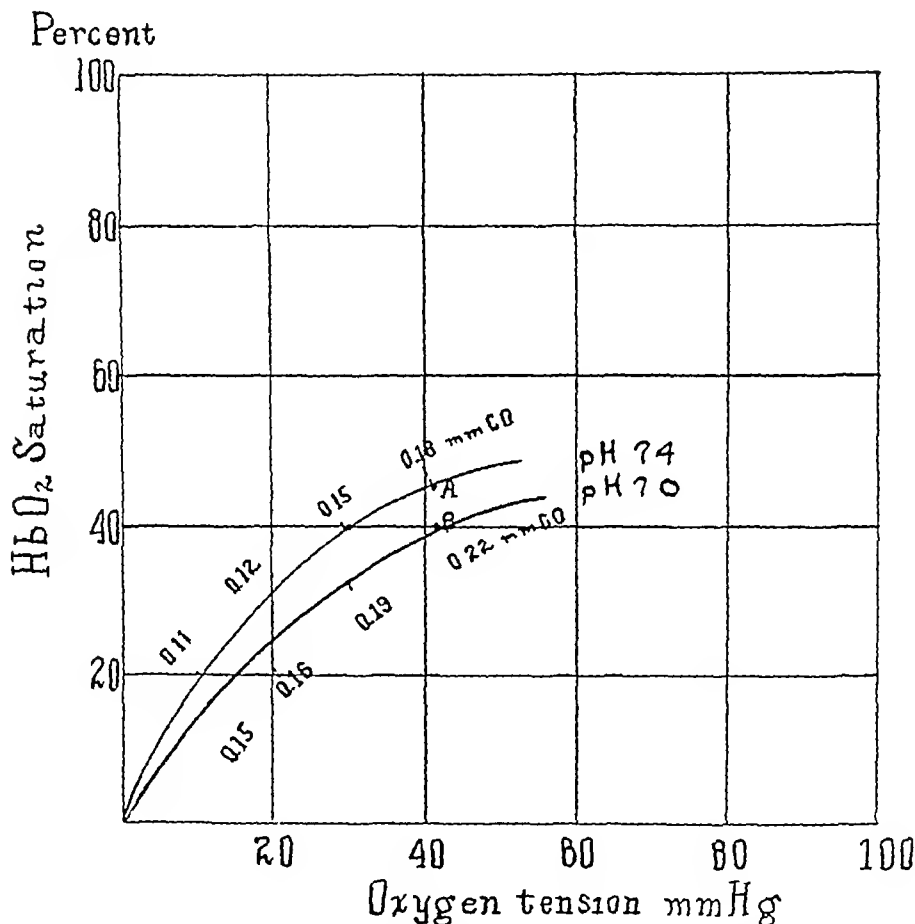


FIG 2 OXYGEN DISSOCIATION CURVE OF BLOOD 50 PER CENT SATURATED WITH CO AND pH 7.4 AND 7.0 SHOWING THE EFFECT OF ACIDITY ON THE PARTIAL PRESSURE OF CO

At constant oxygen tension a lowering of the pH requires an increase of $\frac{0.167 - 0.140}{0.167}$ or 20 per cent in the carbon monoxide tension to maintain the blood at 50 per cent saturation with CO

If, as is the case in the lungs during recovery from asphyxia, the P_{∞} remains constant it is easy to show from equation 5 that the following would be the approximate conditions

$$\begin{aligned} \text{pH} &= 7.0 \\ \text{HbCO} &= 40 \text{ per cent} \\ \text{HbO}_2 &= 40 \text{ per cent} \\ P_{\infty} &= 0.167 \text{ mm Hg} \\ P_{\text{O}_2} &= 30 \text{ mm Hg} \end{aligned}$$

While we have arbitrarily selected a mid portion of the curves for illustration the same relations must necessarily hold true for any and all equilibrium relations, oxygen, carbon monoxide and hemoglobin

TABLE 2

Elimination of CO from partially asphyxiated dogs with constant ventilatory rate and varying acidity of blood

Time minutes	Breathing air		Breathing 10 per cent CO ₂ + Air		HCl intravenously		Hyperventilation	
	HbCO satn. per cent	pH	HbCO satn. per cent	pH	HbCO satn. per cent	pH	HbCO satn. per cent	pH
0	73	7.40	95	7.38	91	7.34	80	7.58
15	55	7.42	57	7.13	48	7.02	57	7.50
30	49	7.40	44	7.08	35	6.94	47	7.48
60	37	7.38	22	7.08	21	6.92	35	7.53
120	28	7.48	6	7.00	10	6.80	23	7.50
Oxygen capacity, volumes per cent	15.7		21.93		21.6		22.6	

It is at once evident from the above that increasing acidity of the blood from whatever cause hastens the elimination of carbon monoxide from the blood independently of ventilatory rate

To test this deduction the following experiments on asphyxiated dogs were done. The results are recorded in Table 2

The rate of elimination of CO from the blood was studied under the following conditions

1 Animal breathing air with constant ventilatory rate. Blood pH normal

2 Blood pH decreased by breathing 10 per cent CO₂ with constant ventilatory rate

3 Giving dilute hydrochloric acid intravenously with constant ventilatory rate

4 Hyperventilation with air

This experimental portion of the work was made possible by the Department of Surgery placing at our disposal their laboratories. We are indebted to Dr. Robert Kapsinow for his kindness in performing the animal surgery necessary.

EXPERIMENTAL

Medium size dogs were used throughout. Operative technic, anesthesia, gassing, blood sampling and blood analysis were the same in all of the experiments. Except in experiment 4 the ventilatory rate was the same throughout the series.

Anesthesia 30 to 40 cc of paraldehyde was given by stomach tube three quarters to one hour before the experiment was started. This was found to be sufficient to obtain complete relaxation for two to three hours.

Operation After the animal was completely anesthetized it was placed on the operating table and a trachea cannula was inserted and connected to an artificial respiration machine. The thoracic cavity was opened by making an incision to the left of the sternum and removing portions of three ribs over the heart. A flap was turned back which gave an opening over the heart of about 10 by 14 cm. Respiration was controlled by an artificial respiration apparatus. Great care was taken to prevent loss of blood. The right femoral artery was exposed for blood sampling. The mediastinal partition was punctured. This caused collapse of the lungs. The rate and degree of expansion and contraction of the lungs were set and maintained at a constant throughout experiments 1, 2, 3 by means of a motor driven artificial respiration machine. The pulmonary ventilation was set at about the normal minute volume and was totally independent of the animal's own respiratory effort.

Gassing The animal was given pure illuminating gas through the artificial respiration apparatus at 15 to 20-second intervals until the blood became about 60 to 80 per cent saturated with carbon monoxide. After a little experience the desired saturation could be estimated by the animal's heart action.

Blood sampling Five to 10 cc samples of blood were drawn from the right femoral artery at intervals with a 10 cc Luer syringe, and delivered into ovalated sampling tubes under oil.

Blood analysis Oxygen capacity, oxygen and carbon dioxide content, carbon monoxide content were determined by the Van Slyke (1924) method. The hydrogen ion concentration of the blood was determined colorimetrically by Cullen's method (1923). The results are given in tables 1 and 2 and figure 1.

In experiment 1 it was found that the blood pH fell slightly during the first half hour of the experiment. To eliminate this complicating factor 10 gm of NaHCO_3

was given intravenously. This was found to maintain the blood pH within normal limits.

DISCUSSION

For simplicity of comparison we have plotted the data of table 2 in figure 3, the initial saturation with CO being the same.

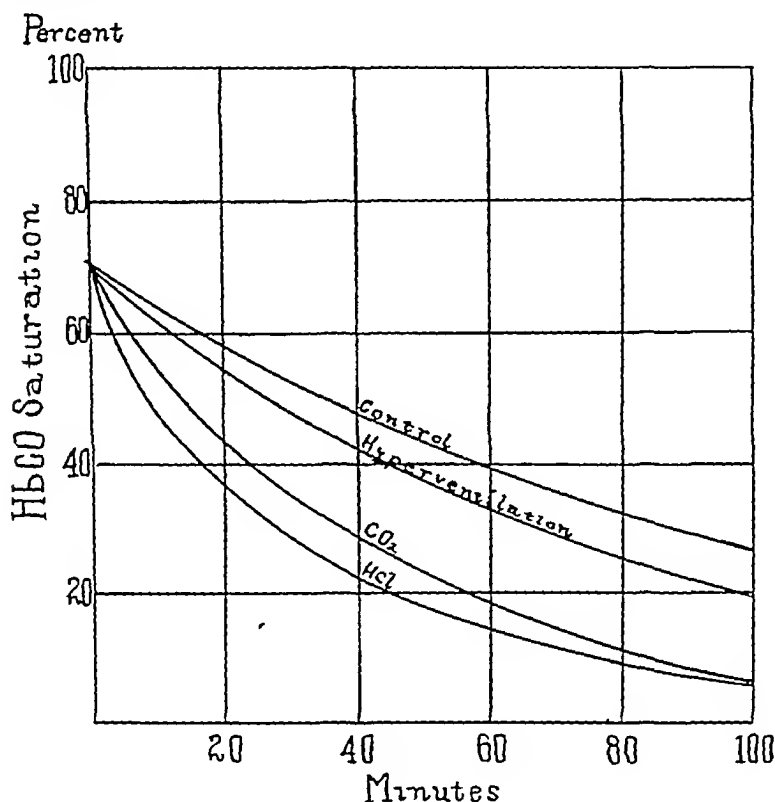


FIG 3 CURVES SHOWING THE RATE OF ELIMINATION OF CARBON MONOXIDE FROM THE BLOOD OF PARTIALLY ASPHYXIATED DOGS UNDER DIFFERENT CONDITIONS

In all cases except one (hyper-ventilation) the ventilatory rate was the same.

The rate of elimination of CO at any instant is proportional to the concentration of CO in the blood, i.e.,

$$\frac{dS}{dt} \propto S$$

where S is the per cent of saturation of blood with CO Integrating

$$\log \frac{S_i}{S_t} = kt$$

Where S is the initial saturation and S_t the saturation at time t If the data of table 2 be plotted using $\log \frac{S_i}{S_t}$ as ordinates and t as abscissae straight line should result This is exactly the case The slopes of the lines give k for the four different conditions and with these k values figure 2 is calculated This allows one to start the curves from a common point and bring out the differences in a striking fashion

Little comment is needed It is clear that hyperventilation itself is of minor importance in the elimination of CO Decreasing pH by any acid, CO_2 or HCl causes a great increase in the rate of elimination and is the most important factor

It must be strongly emphasized, however, that CO_2 administered to asphyxiated subjects has a powerful stimulating effect on respiration While hyperventilation plays a minor rôle yet it is a distinct one The asphyxiated subject has usually a decreased pulmonary ventilation This slows CO elimination Any means of increasing ventilatory rate is beneficial CO_2 has a double function through the same mechanism It increases blood hydrogen ion concentration which in itself favors CO elimination This double function makes it the ideal therapy to employ the addition of pure oxygen while helpful is far less important

The alteration in the oxygen dissociation curve by CO as the cause of the anaemia

An old experiment by Haldane (1895) would force us to believe that CO by itself is non-poisonous He placed mice in a chamber with sufficient CO to saturate completely the hemoglobin It could no longer serve to transport oxygen to the tissues and the mice would have died promptly (as did the controls) had it not been for the fact that the chambers also contained oxygen under several atmospheres of pressure Under these conditions 3 to 4 volumes per cent of oxygen could be carried by the blood in physical solution This was sufficient for the needs of the animals and they remained perfectly well While

it is not entirely clear that these results can be carried over to human beings, since the symptoms of CO poisoning are somewhat anomalous as pure symptoms of anoxemia, yet tentatively we may accept the hypothesis that CO poisoning causes harm solely by depriving the

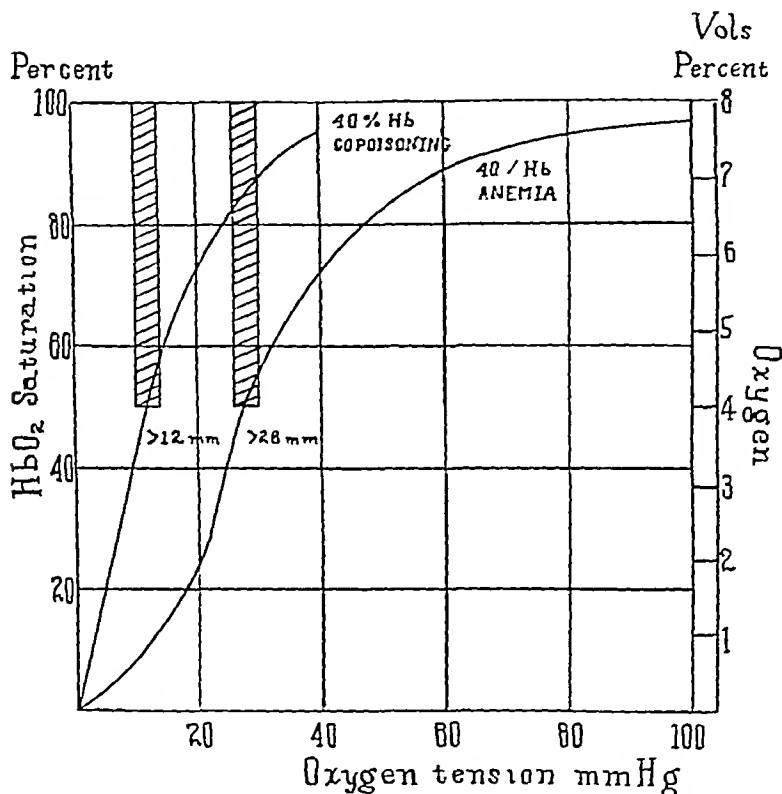


FIG 4 OXYGEN DISSOCIATION CURVE OF THE BLOOD OF A CASE OF CARBON MONOXIDE POISONING 60 PER CENT SATURATED WITH CO CONTRASTED WITH THE CURVE OF A CASE OF PERNICIOUS ANEMIA WITH THE SAME AMOUNT OF AVAILABLE OXYHEMOGLOBIN, VIZ, 40 PER CENT OF NORMAL

tissue of oxygen. There is current, however, a misconception of the mechanism of this anoxemia which figure 4 makes clear. By means of equation 5 we have calculated the oxygen dissociation curve of blood when constantly saturated 50 per cent with CO. The form of this curve so calculated is in complete accord with the experi-

mentally determined curve for mouse blood under the same conditions as reported by Haldane (1912) Forty per cent of the total hemoglobin is available for combination with oxygen and the saturation of this residue of hemoglobin with oxygen is plotted against oxygen tension In contrast the oxygen dissociation curve of a case of pernicious anemia having 40 per cent of the normal amount of hemoglobin is plotted The curve in the latter case is unaltered as one of us has determined³ but the CO case shows a marked alteration in the shape and position of the curve It is this alteration which causes the anoxemia with its train of profound acute symptoms in marked contrast to the relatively normal condition of the anemia case In both cases the maximum amount of oxygen available is the same, 8 volumes per cent In both cases, the physiologic needs of the tissues is the same, say about 4 volumes per cent or half the maximum available For the anemic case, however, this physiologic oxygen requirement (indicated by the hatched column) is dissociable at a partial pressure greater than 28 mm of oxygen Evidently at or above this pressure tissue metabolism is normal

But in the case of CO poisoning, the giving up of 4 volumes of oxygen requires descent to a partial pressure of 12 mm of oxygen Indeed three-fourths of the needed oxygen is dissociated between 28 mm Hg, the minimum for the anemic case, and 12 mm Now it is quite conceivable that the partial pressure at which oxygen is available in the capillaries for tissue metabolism is important Krogh has pictured a single capillary as supplying a cylinder of tissue In order adequately to supply the periphery of the cylinder with oxygen, the oxygen pressure at the capillary wall cannot fall below a certain minimum or else the rate of diffusion of oxygen to the periphery which varies directly as this pressure head will fall below the physiologic requirement Presumably this partial pressure lies somewhere between 12 and 20 mm of Hg approximately

Despite the presence in the blood of two or three times the amount of oxygen necessary for normal tissue function profound anoxemia results in CO poisoning This is due to an alteration of the shape and posi-

³ Stadie Unpublished data

tion of the oxygen dissociation curve causing a marked lowering of the partial pressure at which oxygen is available for tissue metabolism

SUMMARY

An equation is derived showing the relation between hemoglobin, oxygen, carbon monoxide and pH at equilibrium

Experiments in vitro are given substantiating the equation.

By means of this relation it is possible to predict the effect of increasing acidity on the elimination of CO from asphyxiated animals. It is shown that increasing hydrogen ion concentration in itself will hasten elimination regardless of the ventilatory rate. Experiments on dogs showed this to be so.

It is further shown by the equation that the oxygen dissociation curve of hemoglobin is altered by carbon monoxide. Accepting the hypothesis that carbon monoxide is in itself completely non-poisonous, the altered curve shows that even though twice or three times the physiologic requirements of oxygen are present in the blood there is a marked lowering of the partial pressure of oxygen in the capillaries. This conceivably depresses the rate of diffusion of oxygen to the tissues, thus causing anoxemia.

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OBSERVATIONS ON THE EFFECT OF ANOXEMIA ON THE HEART

I AURICULO-VENTRICULAR CONDUCTION

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(Received for publication August 6, 1925)

INTRODUCTION

Since anoxemia is so frequent a manifestation of many medical conditions, and particularly since in some of these abnormal states its severity may make it a prominent feature of the malady, it is of some interest and importance to inquire in how far it may affect various bodily functions. Is anoxemia to be considered merely a symptom, relatively unimportant in itself, except in indicating that the process which causes it may be a severe one, or is it to be viewed in the light of a symptom which, by virtue of its own effects, may have a serious influence on the course of the underlying condition? It is needless to point out that an impaired oxygen supply may have harmful effects on certain physiological activities, the general nature of which has been sufficiently emphasized by Haldane (1919) and Barcroft (1920).

Concerning the influence of anoxemia on the heart not a great deal is definitely known. I do not propose to enter into a detailed review of all the literature bearing on the changes in the heart produced by anoxemia, but shall confine myself particularly to the work which is directly related to the problem of the effect of anoxemia on auriculo-ventricular conduction.¹ Lewis and Mathison (1910-11) first proved

¹ In this paper, I shall use the term "conduction" only to express the transmission of an impulse from the auricles to the ventricles, without reference to the hypothetical function "conductivity," or to the mechanism by which alterations in the transmission time are brought about.

that following asphyxiation depression of auriculo-ventricular conduction took place, ultimately advancing to the degree of complete heart block. Since these events occurred after the complete removal of vagal tone, they were ascribed to a direct effect on the myocardium, and Mathison (1910-11) showed that this result was not caused by the accumulation of carbon dioxide, but rather by lack of oxygen. Later, Mathison (1910) found that the administration of a high concentration of carbon dioxide might bring about a certain degree of auriculo-ventricular block, apparently slight. More recently, Greene and Gilbert (1922) observed the effect of a more slowly induced progressive anoxemia, produced by the method of re-breathing and maintained until the animal died usually within a period of 15 to 18 minutes. Carbon dioxide was continually absorbed in the apparatus which they used, and was thus eliminated as a possible factor in the causation of the changes which took place in the cardiac mechanism. Up to a certain critical period, usually when respiration failed, there were practically no changes in the heart except slight quickening of rate and reduction of the P-R interval. Following the "crisis" however, certain alterations in the rhythm of the heart (including conduction defects) did take place, these changes occurring as a result of central stimulation of the vagus nerves by anoxemia. It was only three to five minutes after respirations had ceased and just before the death of the animal that Greene and Gilbert were able to detect a direct action on the heart by anoxemia. Essentially the same phenomena had been noted previously by Haggard (1921) on exposing animals to increasing concentrations of carbon monoxide. Greene and Gilbert reconciled the difference in the results obtained by them and those observed by Lewis and Mathison by assuming that, because of the rapid method of inducing anoxemia used by the latter, the direct effects on the myocardium appeared early in their experiments and corresponded to the late effects which they (Greene and Gilbert) observed in their own more prolonged experiments.

On the basis of the observations of Greene and Gilbert, then, one might infer that even a severe degree of anoxemia had relatively little influence on the myocardium. It seemed, however, that a more accurate idea of the effect of anoxemia on the heart muscle might be acquired if one subjected an animal to a degree of anoxemia, insufficient

to bring about its death, but maintained over a longer period of time. In this way, conditions would simulate more closely those prevailing in the ordinary clinical cases, in which a moderate or severe grade of anoxemia may be present for a comparatively long time. It was possible that the duration of time in which anoxemia was present might be a factor of some importance in determining the effect on the heart muscle. Moreover, it was conceivable that if one increased the rate of the heart during anoxemia, one might bring out a change which was latent in the naturally beating heart. Erlanger and Hirschfelder (1905) and Lewis and Oppenheimer (1910-11) have shown that when auriculo-ventricular conduction was already impaired, an increase in the heart rate exaggerated the degree of block between the auricles and ventricles.

EXPERIMENTAL METHODS

Dogs, averaging between 10 to 15 kg. in weight were anesthetized by the subcutaneous administration of pantopon, 5.5 to 6.0 mgm. per kilogram, followed within 15 minutes by giving 1 gm. urethane per kilogram by stomach tube. A steady and satisfactory anesthesia was produced, the animal remaining quiet and in good condition for as long as five hours, the longest period over which the experiments were carried out. Both vagus nerves were tied, in addition, in all but a few of the experiments full doses of atropine, usually 2 to 5 mgm., were injected intravenously every 15 to 20 minutes. No differences were noted between those animals which received atropine, and those that did not. After the insertion of a cannula in the trachea, artificial respiration was started, using intratracheal insufflation. Usually the pressure distending the lungs was 20 to 30 mm. mercury. After ligating the internal mammary arteries, the sternum was split in the midline, the sides being retracted by weights. In most of the experiments the pericardium was opened and stitched to the sides of the chest wall, and in these electrocardiographic records of the heart were taken from electrodes placed directly on the auricle. The electrodes were small glass tubes filled with a saturated solution of copper sulphate, the ends of the tubes being plugged with a paste of kaolin in normal saline solution. In other experiments, in which the pericardium was unopened, records were taken by lead II, small German silver electrodes being either sewn under the skin or held in position by bandages soaked in saline.

Anoxemia, always of the anoxic type, was produced by administering suitable mixtures of oxygen and nitrogen. Thick-walled rubber tubing leading from high pressure gas tanks fitted with reducing valves were connected to glass tubes inserted through a rubber stopper in a glass flask. The ends of the glass tubes were placed just beneath a layer of water, the gases bubbling through the water before being forced out of a side tube leading to the intratracheal catheter. Connected

with each rubber tube coming from the tanks was a mercury manometer giving the pressure of each of the gases flowing to the mixing flask, and between the mixing flask and the catheter was another manometer registering the pressure in the the lungs. The amount and proportions of the gases and the degree of distention of the lungs were regulated by varying the pressure in each of the systems. The volume of gas flowing to the lungs was about 5000 cc per minute, insuring adequate ventilation.

The auricles were driven at desired rates by a Lewis rotary stimulator, only break shocks being used, the strength of the shock in every instance being well above the threshold value. This apparatus was connected to small fishhook stimulating electrodes which were inserted near the base of the right auricular appendage. The recording electrodes on the heart, when they were used, were placed about 1 cm. away toward the midcaval region, in line with the stimulating electrodes. In some of the experiments, two galvanometers were used, recording simultaneously on the same film, one for the stimuli, the other for the activity of the heart. Owing to a temporary disarrangement of one of the galvanometers, only one could be used in most of the observations. It was found, however, that this method was quite satisfactory, since by using sufficiently a strong stimulating current, the stimuli were clearly shown on the electrocardiographic record.

Measurements of the P-R interval were made with a Lucas comparator, the average of at least three determinations being taken as the figure for a record. The figures do not represent true P-R values since the measurements were made from the sharpest points, not necessarily the beginning of the auricular and ventricular complexes. Since the same points were used in measuring the curves of an entire experiment, the absolute figures are comparable, then, in the same but not in different animals. In most instances, the figures for a record fell within 0.003 to 0.005 second in the records taken by lead II, and within 0.002 to 0.003 second in the records taken directly from the auricle.

The degree of anoxemia was ascertained by determination of the oxygen saturation of the arterial blood (van Slyke and Stadie, 1921), which was obtained from the femoral artery and handled with the usual precautions. At first, the oxygen capacity was determined on every sample. Later, this was not done, for with sufficiently high concentrations of oxygen (50 to 100 per cent) in the gas mixtures, it was assumed (after a few control observations) that the oxygen saturation was 100 per cent.

The usual procedure of an experiment was to give a high oxygen mixture and to record the activity of the heart with the electrocardiograph during various rates of rhythmic stimulation. The time of stimulation was constant before each record was taken. The percentage of oxygen in the gas mixture was then lowered, and after several minutes records were taken at intervals, at approximately the same rates of stimulation that had been used before. Finally, in most cases, the percentage of oxygen was increased to its previous level and a further series of records again obtained.

TABLE 1
Dog 13 *Pericardium unopened, fully atropinized*

Oxygen saturation 96 per cent*				Oxygen saturation 89 per cent Started 12 45			
Time	Rate of stimulation per minute	Auricular response	P R interval seconds	Time	Rate of stimulation per minute	Auricular response	P R interval seconds
11 55	212	1 1	0 077	12 56	212	1 1	0 078
11 58	213	1 1	0 081	12 57	243	1 1	0 081
12 00	264	1 1	0 083	12 58	261	1 1	†
12 17	285	1 1	0 087	12 59	277	1 1	0 085
12 22	323	1 1	0 097	1 00	324	1 1	0 102
12 24	367	1 1	2 1 response	1 01	370	1 1	2 1 response
12 26	428	1 1	2 1 response	1 02	422	1 1	2 1 response
12 28	476	1 1	2 1, 1 1 response alternating	1 04	475	Occasional (dropped) bent	2 1 with 1 1 response every third bent
12 30	516	Occasional (dropped) bent	2 1, occasional 1 1 response	1 06	518	Occasional (dropped) bent	2 1 with 1 1 response every fourth bent
12 32	572	2 1 response (rate 286)	1 1 response	1 08	580	2 1 response (rate 290)	1 1 response
12 34	610	2 1 response (rate 320)	1 1 response	1 10	658	2 1 response (rate 329)	1 1 response
12 36	768	2 1 response (rate 341)	2 1 response (rate 192)	1 12	774	2 1 response (rate 387)	2 1 response (rate 198 5)

* In this table, as in all subsequent tables, the percentage of oxygen saturation refers to the arterial blood

† Accurate measurements could not be made

RESULTS

The results will be presented in three groups, divided arbitrarily according to the degree of anoxemia, mild (arterial oxygen saturation above 85 per cent), moderate (70 to 85 per cent saturation), severe (oxygen saturation below 70 per cent) The effect of anoxemia on

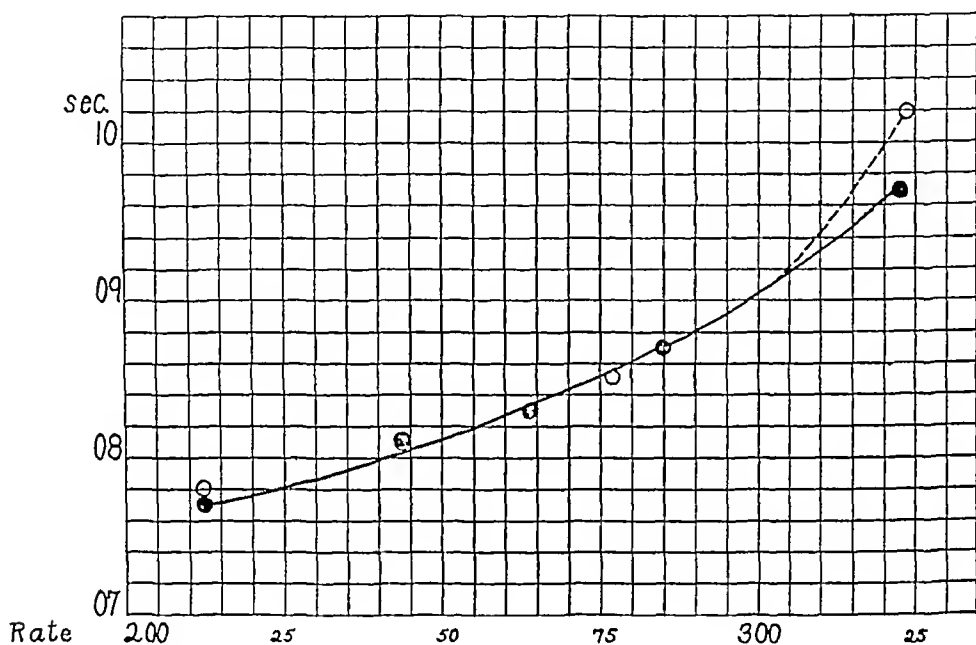


FIG 1 DOG 13 THE P-R INTERVALS AT DIFFERENT RATES OF RHYTHMIC STIMULATION OF THE HEART

Black dots Arterial oxygen saturation 96 per cent

Circles Arterial oxygen saturation 89 per cent

The chart shows the effect of mild anoxemia on auriculo-ventricular conduction. The values for the P-R intervals during the periods of higher and lower arterial oxygen saturation lie on an identical curve, except at the rate of approximately 325 per minute. The apparent divergence of the curves at this point is probably due in part to errors of measurement (see text).

auriculo-ventricular conduction was observed in 17 animals, and since the course of events was essentially the same in the different groups, I shall give the figures only of those experiments in which the data are more complete, or which are of unusual interest.

Effect of mild anoxemia

In tables 1, 2, and 3 are given the results in three experiments. In experiment 13 (table 1), the anoxemia was of relatively short duration.

TABLE 2

Dog 19 Pericardium uropenched, fully atropinized

Oxygen saturation 90 per cent Started 11 40 Natural heart rate 176			Oxygen saturation 98 per cent Started at 12 37 Natural heart rate 164		
Time	Rate of stimulation	P R interval	Time	Rate of stimulation	P R interval
	<i>per minute</i>	<i>second</i>		<i>per minute</i>	<i>second</i>
12 19	221	0 085	1 20	218	0 092
	260	0 092		254	0 100
	312	0 113		312	0 119
12 35	361	Occasional dropped beat	1 28	355	Occasional dropped beat

TABLE 3

Dog 24 Pericardium opened, fully atropinized

Oxygen saturation 100 per cent Natural heart rate 156			Oxygen saturation 89 per cent Started 11 38 Natural heart rate 152			Oxygen saturation 100 per cent Started 1 33 Natural heart rate 144		
Time	Rate of stimulation	P R interval	Time	Rate of stimulation	P R interval	Time	Rate of stimulation	P R interval
	<i>per minute</i>	<i>second</i>		<i>per minute</i>	<i>second</i>		<i>per minute</i>	<i>second</i>
11 15	227	0 092	1 28	223	0 087	2 01	224	0 091
	301	0 107		292	0 101		290	0 104
	342	Rare dropped beats		342	1 1*		338	1 1*
11 29	357	Every sixth or seventh beat dropped	1 32	364	Occasional dropped beat	2 12	358	Occasional dropped beats

* Accurate measurements could not be made

and there was practically no change in the P-R intervals. The values are charted in Figure 1 up to the stage of auriculo-ventricular block.

There is an apparent divergence of the curves at the rate of approxi-

mately 320, but it was more than likely that the difference is due, in part at least, to errors of measurement. One would expect, if there were actually increased impairment of conduction during the anoxic period, that this defect would be enhanced at the more rapid rates. The responses remain, however, practically identical.

In the other two experiments (tables 2 and 3),² a decrease in the

Dog 1.

Oxygen saturation 100 per cent Natural heart rate 162			Oxygen saturation 81 per cent Started 11 47 Natural heart rate 154			Oxygen saturat Started Natural hea	
Time	Rate of stimulation	P-R interval	Time	Rate of stimulation	P-R interval	Time	Rate of stimulation
	<i>per minute</i>	<i>seconds</i>		<i>per minute</i>	<i>seconds</i>		<i>per minute</i>
11 33	204	0 091	11 57	210	0 089	12 30	201
	230	0 091		234	0 091		226
	257	0 096		266	0 095		256
				286	0 099		280
	301	0 099		306	0 102		307
	335	0 116		342	0 113		344
	366	0 125		372	0 123		376
	400	1 1, with periods of 2 1 response		393	1 1, with periods of 2 1 response		399
11 45	438	2 1, with 1 1 response every third beat	12 07	434	2 1, with 1 1 response every third beat	12 40	420

* Accurate measurements could not be made

P-R interval occurred after a somewhat longer period of anoxemia. The difference between the values of the higher and lower oxygen series is slight but definite. It represents a real change, for it occurred uniformly at the different rates of the same series of observations,

² Although a series of records was made during high oxygen concentration, the records were not sufficiently good for accurate measurement. The results are given primarily to show the increase in the P-R intervals following the change from the lower to the higher oxygen saturations of the arterial blood.

and, on restoration of normal oxygen saturation, as in Experiment 24, (table 3), the P-R intervals rose again and approached those originally obtained

Experiments 13 and 24 serve also as controls on the observations to be reported below, for they demonstrate that the mere duration of an experiment produced no significant changes in the P-R interval

Atropinized

Oxygen saturation 81 per cent Started 11 47 Heart rate 156		Oxygen saturation 81 per cent Started 11 47 Natural heart rate 144		Oxygen saturation 97 per cent Started 2-16 Natural heart rate 162		
P R interval	Time	Rate of stimulation	P R interval	Time	Rate of stimulation	P-R interval
<i>seconds</i>		<i>per minute</i>	<i>seconds</i>		<i>per minute</i>	<i>seconds</i>
0 082	2 01	229	0 079	3 01	207	0 077
0 082			0 081		222	0 079
0 084			0 085		257	0 079
0 089			0 091		282	0 090
0 092			0 102		303	0 092
0 100			0 118		330	0 096
0 113			2 1 response		365	*
2 1 response		396			380	0 119
2 1 and 1 1 response	2 15	427	Periods of 2 1, with periods of 1 1 response	3 16	424	Periods of 1 1, with periods of 2 1 response

In experiment 24, after almost three hours, the values at the different rates were practically identical

To summarize, then, the effects of mild anoxemia on auriculo-ventricular conduction there is practically no change over a short period of time, over longer durations, the P-R interval is slightly but definitely lowered at both low and high rates of stimulation. When the normal degree of arterial oxygen saturation is restored, the figures rise again to practically their original values

Effect of moderate anoxemia

Table 4 contains the figures of an experiment in which a moderately severe anoxemia persisted over a long time. The effect was to cause a progressive lowering of auriculo-ventricular conduction time even after two hours of anoxemia. Moreover, after the re-establishment

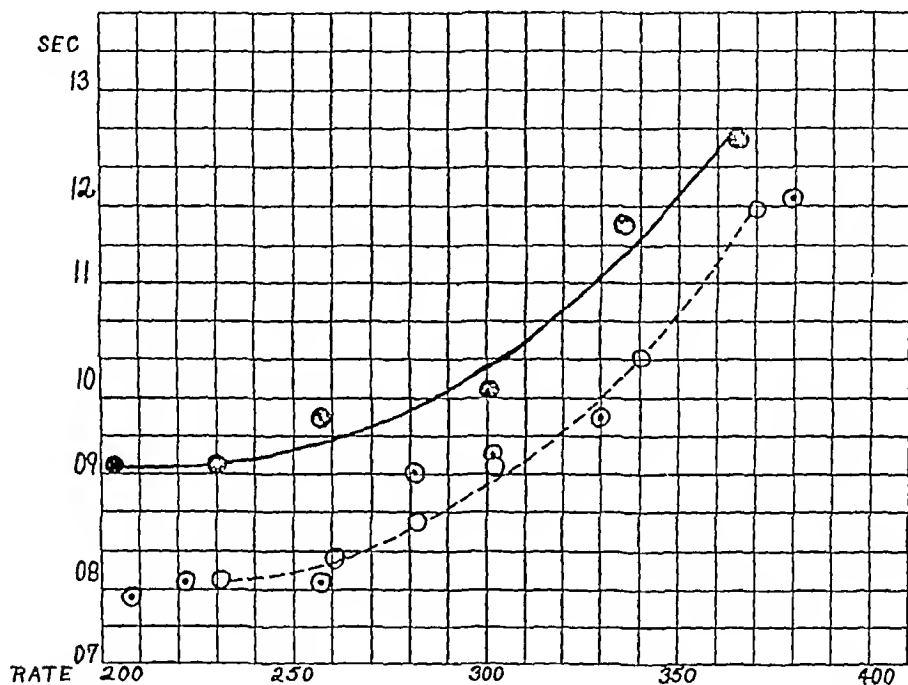


FIG 2 DOG 15 THE P-R INTERVALS AT DIFFERENT RATES OF RHYTHMIC STIMULATION OF THE HEART

The determinations were made in the following order

- 1 Black dots Arterial oxygen saturation 100 per cent (column 1, table 4)
- 2 Circles Arterial oxygen saturation 81 per cent (column 5, table 4)
- 3 Dotted circles Arterial oxygen saturation 97 per cent (column 6, table 4)

This chart illustrates the decrease in the P-R interval during moderate anoxemia, and also the failure of immediate return to the normal figures when normal percentage of arterial oxygen saturation is restored after a long period of anoxemia

of normal arterial saturation (column 6), the figures for the P-R intervals remain essentially at the level of the last series of determinations in the period of anoxemia

Tables 5 and 6 give the summaries of two further experiments. In experiment 22 (table 5), the first series of determinations (column

3) during anoxemia showed a slight fall in the P-R values. In the second series (column 4), the auriculo-ventricular conduction time was still further shortened at the lowest rate of stimulation, as the rate of stimulation rose, however, the P-R interval increased to the normal level, and at the rate of 353 per minute, conduction defects

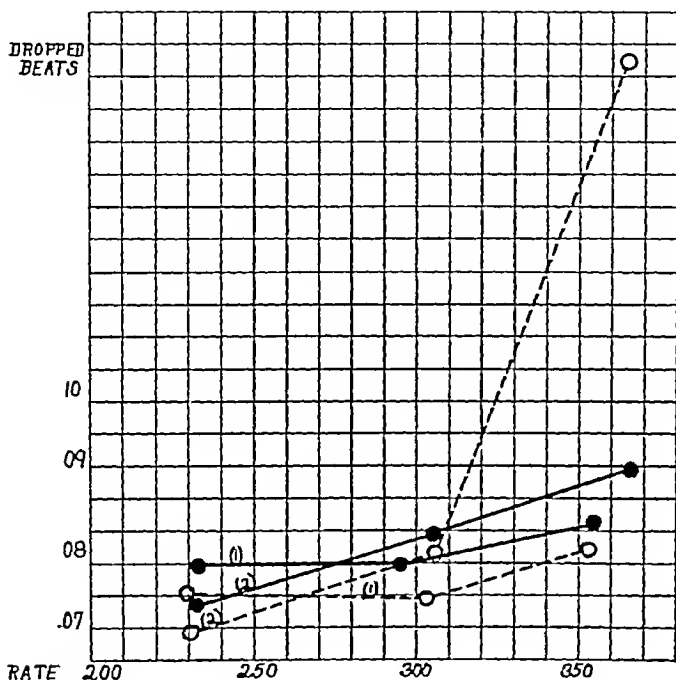


FIG 3 DOG 22 THE P-R INTERVALS AT DIFFERENT RATES OF RHYTHMIC STIMULATION OF THE HEART

The determinations were made in the following order

- 1 Black dots (1) Arterial oxygen saturation 97 per cent (column 1, table 5)
- 2 Circles (1) Arterial oxygen saturation 80 per cent (column 3, table 5)
- 3 Circles (2) Arterial oxygen saturation 80 per cent (column 4, table 5), 15 minutes after the preceding series
- 4 Black dots (2) Arterial oxygen saturation 100 per cent (column 5, table 5)

The chart illustrates the early quickening of conduction during anoxemia, the later stage of impaired auriculo-ventricular conduction, brought out at the rapid rates of stimulation, and the return toward the original P-R values when normal arterial oxygen saturation is restored

TABLE 5
Dog 22 Pericardium opened, fully atropinized

Oxygen saturation 100 per cent			Oxygen saturation 80 per cent Started 11 37			Oxygen saturation 100 per cent Started 12 03			Oxygen saturation 80 per cent Started 12 27			Oxygen saturation 80 per cent Started 12 27			Oxygen saturation 100 per cent Started 1 29		
Time	Rate of stimulation		Time	Rate of stimulation		Time	Rate of stimulation		Time	Rate of stimulation		Time	Rate of stimulation		Time	Rate of stimulation	
	per min- ute	second		per min- ute	second		per min- ute	second		per min- ute	second		per min- ute	second		per min- ute	second
1 19	232	0 078	11 32	230	0 078	12 06	232	0 073	12 39	238	0 076	1 26	226	0 070	1 45	231	0 075
1 21	295	0 078	11 34	300	0 077	12 08	306	0 082	12 40	307	0 077	1 27	302	0 077	1 47	301	0 077
1 24	354	0 083	11 35	351	0 083	12 10	368	0 090	12 42	363	0 082	1 28	354	0 079	1 50	354	0 083

* Every fourth or fifth beat dropped

were definite, there were dropped ventricular beats. On return to the normal oxygen saturation, the figures tended to approach the normal again. In this animal anoxemia was again produced, this time bringing about only the stage of quickened conduction. Again, with the establishment of high arterial oxygen saturation, the figures became normal. In figure 3, the results of this experiment are shown graphically.

I am unable to account for the fact that in the first period of anoxemia, the stage of impaired conduction was brought about in slightly less than one-half hour, whereas later the same degree of anoxemia called forth only the initial stage of quickened conduction after one hour. The conditions of the experiment were apparently unchanged.

TABLE 6

Dog 16 Pericardium unopened, fully atropinized

Oxygen saturation approximately 96 per cent Natural heart rate 130			Oxygen saturation approximately 70 to 75 per cent Started 1 12 Natural heart rate 97		
Time	Rate of stimulation	P R interval	Time	Rate of stimulation	P R interval
	<i>per minute</i>	<i>second</i>		<i>per minute</i>	<i>second</i>
12 50	219	0 092	1 20	216	0 115
	271	0 099	1 25	271	2 1, 1 1 response
	323	0 138			
1 10	372	Periods of 1 1 and periods of 2 1 re- sponse			

In experiment 16 (table 6), only the stage of impaired conduction was seen, without the preliminary period of quickened conduction. Moreover, this change was produced in a very short time. The explanation of this result undoubtedly lies in the fact that during the operative procedure a considerable amount of blood was lost, estimated at one-third to one-half the total blood volume. The animal was in poor condition and, subjected to the stress of both acute hemorrhage and anoxemia, died shortly after the last record was taken at 1 25 p.m.

One may summarize the results obtained in this group of experiments by stating that moderate anoxemia first causes a decrease of the P-R interval. If the anoxemia is carried on for a sufficiently

long time, the stage of impaired conduction appears, and this impairment is most conspicuous at the higher rates of stimulation³ If after a period of anoxemia, normal oxygen saturation of the arterial blood is restored, there is usually a return toward the normal P-R values for the different rates. However, in some instances the recovery may not be complete even after a considerable length of time, the state of auriculo-ventricular conduction may remain very much in the condition in which it was during the last part of the preceding period of anoxemia. The effect of anoxemia is enhanced and hastened if the general condition of the animal is poor.

Effect of severe anoxemia

Tables 7 to 10 show the results of subjecting animals to marked degrees of anoxemia. The results are quite similar to those described in the preceding group except that they appear earlier. In experiment 9 (table 7), in which anoxemia was present for a brief time, only the stage of quickened conduction is seen.

In experiment 12 (table 8), anoxemia was maintained for a slightly longer period but still of relatively short duration. Here, there appeared within a few minutes the stage of quickened conduction. In the next series of records, however, taken but a few minutes later, the figures for the P-R interval were beginning to rise, and the stage of impaired conduction was undoubtedly making its appearance. Unfortunately, a further series of records during anoxemia was not taken.

The extreme effects of anoxemia are shown by experiment 20 (table 9). Here, a very marked grade of anoxemia produced from the first increased P-R intervals. These defects were markedly exaggerated during the rapid rates of beating, especially after the anoxic state had persisted for a time, still relatively short. It is interesting that after return to normal oxygen saturation of the arterial blood, two series of records showed practically no recovery. Figure 4 is a chart of this experiment, and it illustrates well the steepness of the curve of P-R intervals during severe anoxemia, and the failure of recovery after administration of a high oxygen mixture.

³ In experiments other than those described in detail, conduction defects appeared at the low as well as the high rates of stimulation.

In experiment 19 (table 10), marked anoxemia was produced and maintained until the death of the animal ensued. At frequent intervals, the auricles were driven at a rate of approximately 220 per

TABLE 7

Dog 9 Pericardium opened, vagus nerves tied, fully atropinized

Oxygen saturation 96 per cent			Oxygen saturation 40 per cent Started 1 12		
Time	Rate of stimulation	P R interval	Time	Rate of stimulation	R-P interval
	<i>per minute</i>	<i>second</i>		<i>per minute</i>	<i>second</i>
1 01	307	0 139	1 21	304	0 103
1 09	261	0 109	1 20	270	0 099
1 10	210	0 100	1 18	211	0 095

TABLE 8

Dog 12 Pericardium opened, vagus nerves tied, fully atropinized

Oxygen saturation 100 per cent			Oxygen saturation 62 per cent Started 12 45			Oxygen saturation 62 per cent Started 12 45			Oxygen saturation 100 per cent Started 1 03		
Time	Rate of stimulation	P R interval	Time	Rate of stimulation	P R interval	Time	Rate of stimulation	P R interval	Time	Rate of stimulation	P R interval
	<i>per minute</i>	<i>second</i>		<i>per minute</i>	<i>second</i>		<i>per minute</i>	<i>second</i>		<i>per minute</i>	<i>second</i>
12 27	252	0 090									
12 30	282	0 092	12 48	286	0 088	1 00	279	0 091			
12 33	302	0 095	12 50	300	0 088	1 01	294	0 091	1 14	321	0 092
12 35	349	0 112	12 52	330	0 091	1 02	331	0 096	1 15	348	0 096
12 37	382	Dropped beats	12 54	369	0 104	1 03	380	0 110	1 16	392	0 118
			12 56	416	Dropped beats	1 04	422	Dropped beats	1 17	430	Dropped beats

minute. Here, again, there was first a fall, later a rise in the P-R interval.

To sum up the data of this group of experiments practically the same events took place as had been observed when animals were subjected to moderate grades of anoxemia, the changes, however,

TABLE 9
Dog 120 *Pericardium opened, fully atropinized*

Oxygen saturation 97 per cent Started 12 08 Natural heart rate 178			Oxygen saturation 97 per cent Started 12 08 Natural heart rate 179			Oxygen saturation 45 per cent Started 11 42 Natural heart rate 203			Oxygen saturation 97 per cent Started 12 08 Natural heart rate 178		
Time	Rate of stimulation	P-R interval	Time	Rate of stimulation	P-R interval	Time	Rate of stimulation	P-R interval	Time	Rate of stimulation	P-R interval
	per minute	second		per minute	second		per minute	second		per minute	second
11 32	212	0 058	12 03	222	0 068	12 07	352	0 137	12 39	217	0 064
	234	0 057		249	0 072		326	0 137		241	0 070
				271	0 077		300	0 110		270	0 075
	263	0 064		300	0 110		326	0 137		303	0 111
	292	0 067		352	Numerous dropped beats		352	Numerous dropped beats		322	0 128
11 40	338	0 103	12 01	348	Occasional dropped beat	12 07	352	Occasional dropped beats	12 45	358	Occasional dropped beat

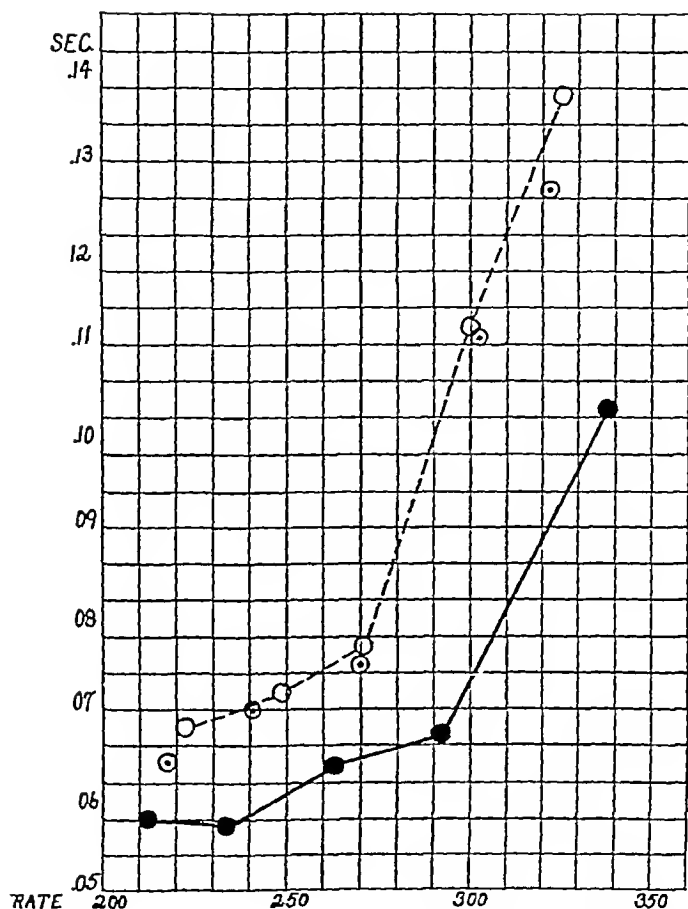


FIG 4 DOG 20 THE P-R INTERVALS AT DIFFERENT RATES OF RHYTHMIC STIMULATION OF THE HEART

The determinations were made in the following order

- 1 Black dots Arterial oxygen saturation 97 per cent (column 1, table 9)
- 2 Circles Arterial oxygen saturation 45 per cent (column 3, table 9)
- 3 Dotted circles Arterial oxygen saturation 97 per cent (column 5, table 9)

The chart shows the effect of severe anoxemia on auriculo-ventricular conduction. The stage of impaired conduction appears early, the impairment being exaggerated at the higher rates of stimulation. The failure of immediate recovery is also well illustrated, the P-R values being almost identical, after one-half hour of normal arterial oxygen saturation, with the values obtained during the anoxic period.

tending to be more marked and to appear more rapidly. In these experiments, too, there was evidence that recovery may be slow after the re-establishment of normal oxygen saturation of the arterial blood.

TABLE 10

Dog 19 Pericardium unopened, fully atropinized

Normal P-R interval at rate of 218 is 0.092 second (oxygen saturation 98 per cent)
 Natural heart rate during normal arterial oxygen saturation is 164 per minute

Time	Oxygen saturation 41 per cent Started 1.28		
	Natural rate	Rate of stimulation	P-R interval*
	<i>per minute</i>	<i>per minute</i>	<i>per minute</i>
1.30	156	218	0.086
1.31.30	158	222	0.084
1.38	157	220	0.086
1.41	158	224	0.087
1.45	161	226	0.086
1.50	163	224	0.088
1.55	155	220	0.089
2.00	152	224	0.090
2.06	135	220	0.094
2.10	114	219	0.109
2.14	108 (Sino-auricular block)	220	Intra-auricular block

* During rhythmic stimulation

DISCUSSION

It is clear that several factors are of importance in determining the effect of anoxemia on auriculo-ventricular conduction. These are the severity and duration of the anoxemia, the rate at which the heart beats and the condition of the animal. In general, anoxemia produces first a decrease, and later an increase of the P-R interval, the latter being favored by severe anoxemia of long duration, by a rapid cardiac rate, and by deterioration of the general condition of the animal, from one cause or another. I have not investigated the influence of alterations of blood pressure. However, Mathison (1910-11) has shown that variations in blood pressure exert no direct effect on the occurrence of heart block in

asphyxia but that an originally high pressure causes auriculo-ventricular block to appear earlier. This result he ascribed to the more vigorous action of the heart and the more rapid exhaustion of the available oxygen supply. This would also appear to be the explanation for the exaggerated effect of anoxemia at high rates of stimulation for under these conditions the work of the heart is increased. It is conceivable that the increases in conduction time at gradually advancing rates of stimulation even during normal arterial oxygen saturation may be explained on the same basis. Barcroft, Bock and Roughton (1921-22) found that in a clinical case of paroxysmal tachycardia there was a striking diminution of the circulatory minute volume associated with an anoxemia of the stagnant type. Since the mean arterial pressure was only slightly decreased, the result of the tachycardia in this case was to increase the work of the heart, at the same time decreasing the total blood supply, and, presumably diminishing the oxygen supply to the heart, to some extent at least. If the lengthening of the P-R interval at increasing cardiac rates is due, however, to a gradually diminishing oxygen supply to the heart, one should be able to find in a series of records taken at different rates a stage in which the conduction time is shorter than it is at still lower rates, since the first effect of anoxemia is to quicken conduction. This phenomenon is not apparent in any of my experiments, that is to say, there has not been an instance in which the P-R interval shortened definitely on increasing the rate of stimulation. The absence of a stage of quickened conduction does not prove, however, that it does not exist, for owing to the fact that the initial rates of beating were relatively high on account of the removal of vagal tone, rates which were slow enough to show such a condition may not have been obtained.

The cause of impairment of auriculo-ventricular conduction in the later stages of anoxemia is definite. It is due to a direct influence on the heart muscle produced by oxygen lack, whatever may be the fundamental mechanism by which such a lack of oxygen exerts its effects. Concerning the cause of the initial quickening of auriculo-ventricular conduction, there may be some question. Vagal influence may be dismissed as a factor in the production of this change in these experiments. However, one must consider the possibility of stimula-

tion of the sympathetic nerves, for such an action is known to hasten auriculo-ventricular conduction. There is a certain amount of contradictory evidence concerning the rôle of the sympathetics in the changes in the heart caused by asphyxia, the effect being judged by alteration in the rate of the heart. Although there is some evidence indicating that the sympathetic nerves may be stimulated during asphyxial or anoxic states, there is other evidence demonstrating that an increase in heart rate may occur, when there is no possibility of sympathetic stimulation. Thus, Greene, Payne, and Siddle (1925) have found, in animals in which the vagus and sympathetic nerves to the heart have been cut, and in which the adrenals have been removed, that although with progressive anoxemia an increase in heart rate does not usually take place, a rise *may* at times occur. Moreover, Mathison (1910) has shown that in the spinal animal cardiac acceleration ensues during asphyxia even after the removal of the upper part of the cord, from which the cardiac accelerator nerves arise.

In the experiments which I performed, the following data favor the view that the decrease in the P-R interval during anoxemia was not entirely due to sympathetic stimulation. (1) The decrease of the P-R interval was not associated with those alterations of the electrocardiogram (when Lead II was used) which have been described by Rothberger and Winterberg (1910). (2) In order to obtain more definite information, I removed in one animal both stellate ganglia, in addition to tying the vagus nerves and administering full doses of atropine. In this animal, as in the others, a decrease in the P-R interval followed the administration of low oxygen mixtures.

The shortening of conduction time was definite, pointing to the conclusion that the change is due to a direct effect on the myocardium.

In view of the work of Hilton and Eichholtz (1925), it is not altogether surprising that a fairly severe and prolonged anoxemia was necessary to bring out the stage of impaired conduction. On subjecting heart-lung preparations of dogs to marked grades of anoxic anoxemia, they found that coincident with the lowering of oxygen tension of the blood, dilatation of the coronary vessels took place, so that the blood supply to the heart was increased. This augmented blood supply was usually able to compensate for the diminished amount

of oxygen available from each unit of blood to such an extent that the oxygen consumption of the heart remained normal. In some instances, however, there was evidence of an oxygen debt, that is to say, the oxygen requirements of the heart could not be fully met. At least one other factor besides the dilatation of the coronary vessels is of importance in compensating for a diminished arterial oxygen tension. The increased hydrogen ion concentration of the blood which was probably present, shifts the oxygen dissociation curve considerably to the right, the result being that at low tensions of oxygen (as would be present in the tissues) a liberation of oxygen that is greater than normal occurs.⁴

It must be remembered that while the circumstances which called forth impaired auriculo-ventricular conduction were severe, and are hardly ever encountered clinically, these observations were made on animals with normal hearts. These experiments show, indeed, that the normal myocardium can be subjected to a degree of anoxemia such as one ordinarily sees clinically, for a short period of time at least, without conspicuous change of conduction from auricles to ventricles.

⁴It is unnecessary to describe in detail the work which has been done on the pH of the blood in anoxemia. This has been adequately done by Koehler, Brunquist, and Loevenhart (1925) who showed in their own investigations that the pH of the blood during anoxemia depends on the balance of two factors: excessive ventilation tending to decrease the hydrogen ion concentration, accumulation of acid products, formed in the tissues as a result of impaired oxidation, tending to increase the acidity of the blood. They reached the conclusion that although there is an alkalosis at first, this increased pH was due to the greater effect by over-ventilation over the acidotic processes, which, however, occurred from the beginning.

In the absence of pH determinations, I cannot state with assurance the alterations that took place in the hydrogen ion concentration in the blood in my experiments. There was probably, during the periods of normal arterial oxygen saturation, an increased alkalinity of the blood due to the washing out of carbon dioxide by the large amount of gas mixtures flowing through the lungs, the amount remaining practically constant throughout an experiment (about 5000 cc.). Since the animals were unable to vary the amount of ventilation (because intratracheal insufflation was used) and so to compensate for the anoxemia by overbreathing, this element of the two factors determining the pH is eliminated. Consequently, any changes in the pH must have been toward the acid side, and Koehler, Brunquist, and Loevenhart have shown that the acidity may become very great in anoxemia.

On the other hand, it is not unreasonable to assume that under anoxemia a heart which is already damaged or which in addition to anoxemia is laboring under some other strain such as a bacterial infection may exhibit relatively early disturbances of function which appear late in a normal heart. This view is given some support by the results obtained in experiment 16 in which because there had been a severe hemorrhage conduction defects appeared after a short period of anoxemia without the stage of quickened conduction being detected. Additional clinical evidence is afforded by the clinical cases of asphyxial block in which varying degrees of impairment of auriculo-ventricular conduction are present in myocardial failure the conduction defects disappearing with improvement of the circulatory condition.

TABLE II

Dog 31. Vagus nerve tied; vagus arteriotomy, both stellate ganglia removed

Oxygen saturation 100 per cent			Oxygen saturation 85 per cent Started 12 28		
Time	Heart rate	P-R interval	Time	Heart rate	P-R interval
	<i>per minute</i>	<i>seconds</i>		<i>per minute</i>	<i>seconds</i>
12 10	165*	0 105	12 38	166*	0 096
			12 36	312†	0 127
			12 45	312†	0 119

* Natural heart rate

† Rhythmic stimulation

SUMMARY AND CONCLUSIONS

Under the conditions in which the above experiments were performed.

1 Anoxemia (of the anoxic type) brings about first a shortening, then lengthening of auriculo-ventricular conduction time.

2 The effect of anoxemia on conduction is influenced by the degree of anoxemia, the length of time in which the heart is subjected to anoxemia, by the rate at which the heart beats, and by the general condition of the animal. There are undoubtedly other factors of importance which I have not investigated.

3 The appearance of the stage of impaired conduction is favored by the *chronic* grades of anoxemia, acting over a long period of time,

by rapid cardiac rates, and by deterioration of the general condition of the animal

4 The changes in conduction produced by anoxemia are due to a direct effect on the myocardium

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OBSERVATIONS OF THE EFFECT OF ANOXEMIA ON THE HEART

II INTRAVENTRICULAR CONDUCTION

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(Received for publication August 6, 1925)

INTRODUCTION

In a preceding paper (Resnik, 1925), I have described the changes in auriculo-ventricular conduction which occur as a result of anoxic anoxemia. In this paper, I shall record the effects of anoxemia on intraventricular conduction. It is well known that supraventricular impulses may give rise to aberrant ventricular complexes, that is to say, ventricular complexes which tend to have the form seen in bundle branch block, even when there is no actual anatomical lesion of the branches of the main conduction stem (Lewis, 1920). Robinson (1916) among others, has reported interesting clinical examples of this kind of disturbance of cardiac mechanism, and Lewis (1911-12) has described its appearance in asphyxial experiments. This form of abnormal ventricular complex, when it is not due to gross structural change, is usually seen in patients who exhibit other evidences of functional impairment of the heart, frequently appearing during periods of myocardial failure, and disappearing with improvement of the circulation. Ventricular aberration may also appear when an apparently normal heart is subjected to some unusual strain, such as the unusually rapid rate of beating in paroxysmal tachycardia of auricular origin. The explanation given for the presence of the abnormal form of the ventricular complex under these conditions is that because of failure of complete recovery of one or the other of the bundle branches of the conduction system, the excitation wave travels through the ventricles at unequal rates of speed the result being that ventricular excitation is set up in an abnormal fashion.

METHODS

In an attempt to determine in how far anoxemia may be responsible for changes in conduction through the ventricles, I have measured the QRS intervals in the records taken with lead II in the experiments previously reported (Resnik, 1925). The methods of the experiments have been fully described in the above paper, and it need only be mentioned that the pericardium was unopened in all the present experiments, and that the animals were completely under the influence of atropine. Measurements were made with a Lucus comparator, 11, the average of at least three determinations being taken as the figure for a single record. The same points for measurement were used throughout the different records of a single experiment. These points, chosen for accuracy and convenience of measurement, did not necessarily include the entire QRS interval. The figures for a single record usually fell within 0.002 to 0.003 second.¹

RESULTS

The results of four experiments are given in tables 1 to 4.

In experiments 13 and 19 (tables 1 and 2), there was practically no change in the duration of the QRS interval, such differences as there were falling within the limit of error of measurement. In experiment 17 (table 3) one in which anoxemia was severe but of short duration, and in experiment 15 (table 4) in which a moderate anoxemia was maintained for two and a half hours, the change was still slight, but more definite. In the latter experiment, the figures remained essentially the same, on return to normal oxygen saturation of the blood, as those obtained in the last series during anoxemia, a phenomenon which was frequently observed in studying the effect of anoxemia on auriculoventricular conduction.

Although records were made at higher rates of stimulation, measurements of the QRS interval were not made in these, because the change in the form of the ventricular complexes and the fusion of the auricular and ventricular components of the curves, would have made them inaccurate. At these high rates of stimulation, the T wave became

¹ Throughout the text and in the tables, it will be understood that the arterial blood is meant when reference is made to the oxygen saturation of the blood.

TABLE 1

Dog 13

Oxygen saturation 96 per cent				Oxygen saturation 89 per cent Started 12 46			
Time	Rate of stimulation	QRS interval	T wave	Time	Rate of stimulation	QRS interval	T wave
	<i>per minute</i>	<i>second</i>			<i>per minute</i>	<i>second</i>	
11 55	212	0 043	Upright	12 56	212	0 041	Upright
	243	0 043	Upright		243	0 042	Upright
	264	0 043	Upright		261	*	Upright
	285	0 042	Upright		277	0 039	Upright
12 22	323	0 041	Inverted	1 00	324	0 042	Inverted

* Record not satisfactory for accurate measurement.

TABLE 2

Dog 10

Oxygen saturation 90 per cent Started 11 40				Oxygen saturation 98 per cent Started 12 37			
Time	Rate of stimulation	QRS	T wave	Time	Rate of stimulation	QRS	T wave
	<i>per minute</i>	<i>second</i>			<i>per minute</i>	<i>second</i>	
12 19	176	0 043	Upright	1 18	148	0 043	Upright
	221	0 043	Upright		218	0 042	Upright
	260	0 043	Upright		254	0 042	Upright
12 32	312	—	Inverted	1 26	312	—	Inverted

TABLE 3

Dog 17

Oxygen saturation 86.5 per cent				Oxygen saturation 49 per cent Started 11.57			
Time	Rate of stimulation	QRS	T wave	Time	Rate of stimulation	QRS	T wave
	<i>per minute</i>	<i>second</i>			<i>per minute</i>	<i>second</i>	
11 30	222	0 046	Upright	12 05	226	0 042	Upright
	282	0 047	Upright		285	0 042	Upright
11 38	331	—	Inverted	12 09	330	—	Inverted

negative and the ventricular complex diphasic. It is probable that these ventricular complexes, which may be termed aberrant, represented transitional forms between the normal and those due to well

Oxygen saturation 100 per cent				Oxygen saturation 81 per cent Started 11 47				Oxygen saturation Started 1		
Time	Rate of stimulation	QRS	T wave	Time	Rate of stimulation	QRS	T wave	Time	Rate of stimulation	QRS
	per minute	second			per minute	second			per minute	second
11 33	204	0 051	Upright	11 57	210	0 051	Upright	12 30	201	0 0
	230	0 049	Upright		234	0 053	Upright		226	0 0
	257	0 053	Upright		266	0 052	Upright		256	0 0
11 40	301	0 056	Upright		286	0 049	Upright		280	0 0
	335	*	Inverted		306	0 051	Upright		307	0 0
				12 03	342	0 053	Inverted	12 36	344	*

* Record not satisfactory for accurate measurement

developed bundle branch block, in spite of the fact that there was no conspicuous widening of the QRS interval. Wilson (1915) found that the most constant feature of transitional complexes was their diphasic character. I have therefore noted in the tables given above the direction of the T wave. Using this as a criterion, it is again apparent that anoxemia produced no striking changes in intraventricular conduction.

In one experiment, no. 19 (table 5), a severe degree of anoxemia was instituted and maintained until the animal died, the rate of stimulation being approximately 220 per minute for each record.

In this experiment there was a gradual increase in the QRS interval, slight, however, until just before the animal died.

At rapid rates of stimulation there was occasionally observed alternation in the height of the ventricular complexes and in the width of the QRS interval. Thus, in experiment 13, the following figures for successive ventricular cycles were obtained at the rate of 323 per minute: 0 0407, 0 0392, 0 0427, 0 0405, 0 0427, 0 0398, 0 0426, 0 0403, 0 0417, 0 0432, 0 0418. There was no greater tendency for these phenomena to occur in either the periods of low or normal oxygen saturation of the arterial blood.

It may be mentioned that there was a very definite tendency for ventricular fibrillation to develop during anoxemia, particularly when the anoxemia was severe and prolonged. This arrhythmia

Oxygen saturation 81 per cent Started 11 47			Oxygen saturation 81 per cent Started 11 57				Oxygen saturation 97 per cent Started 2 16			
Rate of stimulation	QRS	T wave	Time	Rate of stimulation	QRS	T wave	Time	Rate of stimulation	QRS	T wave
per minute	second			per minute	second			per minute	second	
205	0 050	Upnrght					3 01	207	0 046	Upnrght
220	0 048	Upnrght	2 01	229	0 047	Upnrght		222	0 046	Upnrght
254	0 048	Upnrght		260	0 048	Upnrght		257	0 047	Upnrght
279	0 048	Upnrght		282	0 049	Upnrght		282	0 048	Upnrght
301	0 049	Upnrght		304	0 051	Upnrght		303	0 048	Upnrght
337	0 050	Inverted	2 10	341	*	Inverted	3 10	330	0 050	Inverted

TABLE 5

Dog 19 Normal QRS interval at rate of 218 is 0.042 second (Oxygen saturation 98 per cent)

Time	Oxygen saturation 41 per cent Started 1 28		
	Rate of stimulation	QRS	T wave
	per minute	second	
1 30	218	0 041	Upnrght
1 30 30	222	0 041	Upnrght
1 38	220	0 041	Upnrght
1 41	224	0 042	Slightly inverted
1 45	226	0 042	Slightly inverted
1 50	224	0 045	Slightly inverted
1 55	220	0 044	Slightly inverted
2 00	224	0 045	Slightly inverted
2 06	220	0 047	Slightly inverted
2 10	219	0 063*	Diphasic
2 14	220	Ventricular fibrillation	

* R notched, low voltage

occasionally appeared spontaneously, more often it occurred on the lightest handling of the ventricles

DISCUSSION

These experiments are few in number, but they give a fairly clear idea of the effect of anoxemia on the QRS interval. No definite altera-

tions took place except when the anoxemia was of long duration or of marked severity. These results are similar to those of Lewis (1911-12) who found that, whereas asphyxia produced uniformly profound disturbances in auriculo-ventricular conduction, only occasionally were aberrant ventricular complexes present in the same experiments. Moreover, in his experiments the abnormal ventricular complexes were not typical examples of complete bundle branch block, and occurred, apparently, only in the late stages of asphyxiation.

While the changes are slight, showing that the tissues which transmit the wave of excitation through the ventricles are relatively resistant to anoxemia, they are similar in character to those which occurred in auriculo-ventricular conduction, for shortening of the QRS interval appeared first, but was followed by lengthening later. The alterations in intraventricular conduction seen in these experiments were not nearly so conspicuous as many of the clinical examples which have been reported. Yet, it must be emphasized again, that these observations were made almost always in patients who showed other evidence of myocardial disease. The inference that may be drawn, then, is that when ventricular aberration, independent of organic lesions of the bundles of the main conduction stem, is not due to an unusually rapid heart rate (when it may appear in the normal heart with the arterial blood normally saturated with oxygen) it is an indication of a more or less severe functional impairment of the myocardium. This view is based on the assumption that incomplete recovery of the bundle branches, to which aberration of the ventricular complexes is attributed, is due fundamentally to a defective oxygen supply to the myocardium. The anoxemia of myocardial failure, for example, possibly insufficient in itself to bring about intraventricular conduction defects, makes its effects evident when it becomes engrafted on a latent functional impairment of the Purkinje tissue.

SUMMARY AND CONCLUSIONS

- 1 In the normal heart, anoxemia produces comparatively slight changes in intraventricular conduction.

- 2 These changes are similar in character, though much less

marked than those which occur in auriculo-ventricular conduction at first the QRS interval shortens, later it lengthens

3 It is probable that ventricular aberration in clinical cases is an indication of more or less serious disturbance of the function of the Purkinje tissues

4 Ventricular fibrillation is prone to develop during anoxemia

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OBSERVATIONS ON THE EFFECT OF ANOXEMIA ON THE HEART

III CHANGES IN THE AURICLES, WITH PARTICULAR REFERENCE TO THE RELATIONSHIP BETWEEN ANOXEMIA AND AURICULAR FIBRILLATION

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(Received for publication August 6, 1925)

INTRODUCTION

In an earlier paper (Resnik, 1924), I reported several cases in which transient auricular fibrillation occurred as a result of the administration of digitalis. From observation of the response of these patients to atropine, the conclusion was reached that in some instances the drug acted directly on the myocardium. This was apparently at variance with experimental data of Lewis, Drury, Wedd and Iliescu (1921), who showed that strophanthin, acting directly on the auricle of the dog, *hindered* the development of auricular fibrillation, by increasing the absolute refractory period of the auricle. I suggested, therefore, that possibly the functional changes associated with myocardial failure, in which condition anoxemia is practically constant, may have so altered the auricular muscle as to permit the onset of auricular fibrillation, in spite of the increase in the absolute refractory period by digitalis. Other clinical and experimental observations have been reported which suggest that anoxemia predisposes the auricles to fibrillation. Vaquez (1911) mentions the appearance of transient auricular fibrillation in patients with myocardial failure, the arrhythmia disappearing with circulatory improvement. Transient auricular fibrillation is relatively common in lobar pneumonia, being present in about 3 to 5 per cent of patients with this disease (Cole, 1920), one in which anoxemia is often a prominent symptom, but in which significant heart lesions are not usual (Cohn and Jamieson,

1920) Although it is true that in lobar pneumonia the influence of a toxin is to be considered, I am unable to find that in other types of pneumococcus infections in which the same toxin should be at work, auricular fibrillation develops in the same percentage of cases as is true in lobar pneumonia. Experimentally, de Boer (1921) has shown that in the bled frog's heart, in which, among other nutritional disturbances, lack of oxygen must be assumed to be present, properly timed stimuli may cause the onset of fibrillation, whereas in the normally nourished heart, this arrhythmia does not appear.

With these data in mind, suggestive but not conclusive, since in each of these examples there are other factors besides anoxemia which may play a role, I have attempted to determine more definitely the relationship between anoxemia and auricular fibrillation. I have already pointed out (Resnik, 1925) that ventricular fibrillation is prone to develop when the heart has been exposed to anoxemia.

EXPERIMENTAL METHODS

These have been described in a former communication (Resnik 1925a) and it is only necessary to describe those procedures which have not been mentioned before. In order to determine the relative susceptibility of the auricles to develop fibrillation in periods of normal and low oxygen saturation of the arterial blood,¹ the effect of faradic stimulation of the auricles was observed. In all experiments the vagus nerves were tied, and in all but one (experiment 35) the heart was fully under the influence of atropine. Throughout an experiment, the strength of the stimulating current, always well above the threshold value, was unchanged, as was the duration of each stimulation, usually 15 to 30 seconds.

The method and precautions used in determining the refractory period of the atropinized auricles were those described by Lewis, Drury and Bulger (1921), break shocks were used exclusively as rhythmic stimulating and testing shocks. The timing device of Dieuaide and Turner (1924) was used to facilitate the refractory period determina-

¹ In all further mention of the oxygen saturation of the blood, either in the text or in the tables and figures, it will be understood that it is to the arterial blood that the reference is made.

tions In a few of the earlier experiments, two galvanometers were used simultaneously, one to record the rhythmic and interrupting stimuli, the other to record the responses of the auricular muscle. Later, only one galvanometer was available. It was found that with a sufficiently strong current, the signals were plainly visible on the film recording auricular activity, so that satisfactory measurements were possible by using one galvanometer alone, those records only from which accurate determinations could be made being used.

RESULTS

Effect of anoxemia on rate of impulse formation at the sino-auricular node The results which I obtained were in agreement with those that have been observed by others, for there was first an acceleration

TABLE 1
Dog 34 Vagus nerves tied, fully atropinized

Time	Oxygen saturation	Natural rate of heart
	<i>per cent</i>	<i>per minute</i>
11 01	100	205
11 26	90	
11 32	90	212
11 46	90	206
11 53	90	197
12 03	90	185
12 03	90	126

and then a depression of the natural rate of the heart. Table 1 is illustrative of the response of the sino-auricular node to anoxemia.

Following a slight acceleration, the rate of the heart decreased gradually as anoxemia persisted until the condition of the animal was impaired, when the rate slowed rapidly. On one occasion so-called sino-auricular block was produced. The period of acceleration was usually of brief duration and the stage of depression was generally seen at the time when auriculo-ventricular conduction was quickened. This difference in the behavior of the sino-auricular and auriculo-ventricular tissues is shown in table 2.

When anoxemia was relatively mild re-establishment of the normal oxygen saturation of the blood was associated with a return of the heart rate to the normal level. When anoxemia was more severe

and prolonged, however, the rate tended to remain low even when other functions, such as auriculo-ventricular conduction, became normal. In other words, the sino-auricular node is apparently highly sensitive to anoxemia; the stage of depression appears rapidly and recovery is slow.

I have discussed elsewhere (Resnik, (1925a)), the question of stimulation of sympathetic nerves in anoxemia. Although the acceleration in cardiac rate may, in part, be due to this, it seems clear that stimulation of sympathetic nerves does not entirely account for the increased heart rate. In one animal (experiment 31), in addition to tying both vagi and administering full doses of atropine, both stellate ganglia were removed. The heart rate was 163 per minute when the arterial

TABLE 2
Dog 15 Vagus nerves tied, fully atropinized

Time	Oxygen saturation	Natural rate of heart	P R interval
	<i>per cent</i>	<i>per minute</i>	<i>second</i>
11 33	100	162	0 080
11 47	81		
11 57	81	154	0 075
12 30	81	158	0 073
1 15	81	156	0 072
2 01	81	144	0 067
2 16	97		
3 01	97	162	0 070

blood was completely saturated with oxygen, fourteen minutes after a moderate anoxemia (oxygen saturation 85 per cent) was established, the rate rose to 172 per minute. The increase must have been due to a direct effect of anoxemia on the myocardium.

Faradic stimulation of the auricles during anoxemia. It is well known that under ordinary conditions faradic stimulation of the auricles leads almost constantly to the development of auricular fibrillation. If atropine is previously administered, however, in doses sufficient to paralyze the vagal endings, it is difficult, and sometimes impossible, owing to the prolongation of the absolute refractory period of the auricles by this drug, to produce auricular fibrillation by faradic stimulation (Winterberg, 1908). In eleven experiments I have compared the effect produced during periods of normal and low

saturations of oxygen, of faradic stimulation of the auricles. In ten of these observations, vagal tone was completely removed, by tying both vagus nerves and giving frequent large doses of atropine. In one further experiment (experiment 35) atropine was not given, and under these conditions it may be assumed that vagal influence was largely but not entirely removed, since the vagal nerve endings were capable of being stimulated by the faradic current.

A strong faradic current was thrown into the auricles, the stimulating electrodes being placed at or near the junction of the superior vena cava with the upper end of the body of the auricle, and the duration of the after-effect was noted. This procedure was carried out a number of times, so that a clear idea of the susceptibility of the auricles to fibrillate could be obtained. If fibrillation² resulted and lasted for more than one and a half to two minutes, or sometimes longer, faradic stimulation was employed to bring the circus movement to an end. This was done in order that more numerous observations could be obtained.

Using this method, I found that in six of the eleven experiments, there was a distinctly greater tendency for fibrillation to appear after the cessation of faradic stimulation during anoxemia than during normal oxygen saturation of the blood. In one experiment,³ evidence to the same effect was suggestive but not conclusive. In three further experiments there was no preponderating tendency in either anoxemia or normal oxygen saturation of the blood for auricular fibrillation to develop. Finally, in one experiment, there was a greater tendency for fibrillation to develop in the period of normal oxygen saturation of the blood than during anoxemia.

In table 3 are given the data of an experiment which illustrates

² In this connection, I shall mean "fibrillation" to express both fibrillation and flutter, since the work of Lewis and his associates has demonstrated that both these types of arrhythmia have a common underlying mechanism.

³ Two other experiments may be classed as being suggestive of the influence of anoxemia in predisposing the auricles to fibrillation. Thus, in one, no after-effect was observed after 25 stimulations with a faradic current. When anoxemia was instituted, fibrillation appeared after the third faradic stimulation and lasted for five minutes. No further attempts were made to note the effect of faradic stimulation, as other information was being sought.

the type in which anoxemia favored the development of auricular fibrillation

In this experiment, ten trials with the faradic current failed to produce auricular fibrillation when the blood was normally saturated with oxygen. Severe anoxemia was started, shortly afterwards, following three out of four faradic stimulations, fibrillation developed. Each period of fibrillation lasted about three minutes and ended when further faradic currents were thrown into the auricles. On restoration of normal oxygen saturation of the blood, three further trials failed to produce auricular fibrillation.

In table 4 is summarized another experiment of the same kind. It is given in greater detail, since it exhibits certain features which are worthy of mention.

TABLE 3

Dog 8

Oxygen saturation	Number of faradic stimulations	Number of after-effects
<i>per cent</i>		
90	10	0
33	4	3*
91	3	0

* Each of these lasted about three minutes and was ended by faradic stimulation

In this experiment, as in the one previously described, there was a definitely greater tendency for after-effects to appear during anoxemia than during the first period of normal oxygen saturation of the blood. When a high oxygen mixture was given following the preceding anoxemia, the auricles continued to exhibit for a short time a predisposition to fibrillation, which, however, disappeared later. That is to say, immediately after the cessation of anoxemia, the auricles behaved as if they had not fully recovered, and this I have observed on several other occasions.

Experiment 35 (table 5) illustrates another point of interest. The vagus nerves of the animal were tied, atropine was not used. The after-effects are classified in the table as short (5 to 59 seconds) and long (more than one minute). Fifty trials with faradic stimulation, five times associated with vagal stimulation, were ineffective in causing fibrillation during the first period of normal arterial oxygen

TABLE 4

Dog 10

Oxygen saturation	Faradic stimulation	Duration of after-effect
<i>per cent</i>		<i>second</i>
94.5	1	0*
	2	0
	3	0
	4	0
	5	1 minute†
	6	0
	7	0
	8	0
	9	0
	10	0
85	1	30
	2	0
	3	20
	4	15
	5	15
	6	3 minutes†
	7	0
	8	10
	9	0
	10	2 minute
	11	0
96	1	1 minute
	2	0
	3	0
	4	3 minutes†
	5	0
	6	0
	7	10
	8	0
	9	0
	10	0
	11	0
	12	0

* After-effects of less than five seconds duration are recorded as 0

† Ended by faradic stimulation

saturation. A moderately severe anoxemia was produced and in eight attempts, all without vagal stimulation there resulted one

short and one long after-effect. On return to a high oxygen mixture, five attempts, three with vagal stimulation, failed to bring on fibrillation. Anoxemia was again started, and this time five trials, associated with vagal stimulation, caused an after-effect to appear in each instance, the last one, of 44 minutes duration, ceased when normal oxygen saturation of the blood was restored. At this time, with a high oxygen mixture, two of four faradic stimulations, each combined with vagal stimulation, produced auricular fibrillation. Finally, when a severe anoxemia was started, seven trials failed to cause an after-effect.

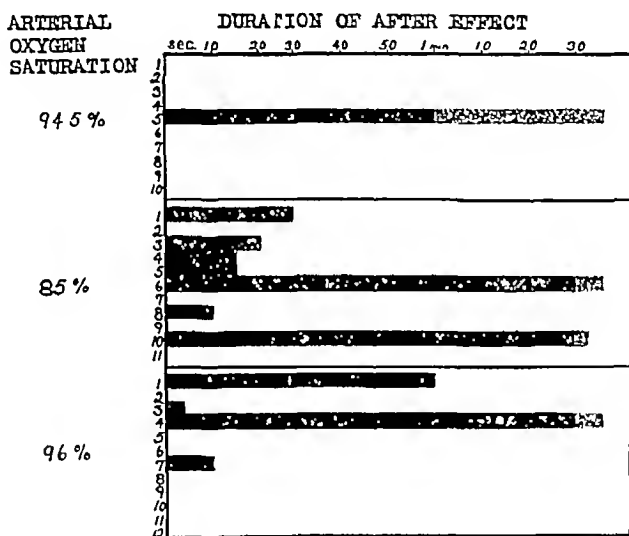


FIG. 1. EXPERIMENT 10. THE RESPONSE OF THE AURICLES (VAGAL TONE COMPLETELY REMOVED) TO FARADIC STIMULATION DURING PERIODS OF NORMAL AND LOW OXYGEN SATURATION OF THE BLOOD.

The chart is divided into three parts, the upper representing 10 trials with faradic stimulation, the arterial oxygen saturation being 94.5. The middle part shows the result of 11 faradic stimulations, the blood oxygen saturation being 86. The lower part shows the effect of 12 stimulations when the oxygen saturation of the blood was raised again to 96. The black bars represent the duration of the after-effect, the stippled areas indicating that fibrillation lasted longer than the length of the black bar indicates and was ended by stimulating the auricles with a faradic current (except no. 10 of the middle block which ended spontaneously in two minutes). Note the increased tendency for the auricles to fibrillate during the anoxic period, and also the persistence of this tendency for a short time after the normal oxygen saturation of the blood was restored.

This experiment demonstrates again the effect of anoxemia in predisposing the auricles to fibrillation. It shows also that following a period of anoxemia, the recovery process in the auricles is delayed, so that fibrillation may be produced with the blood fully saturated with oxygen, whereas previously under the same conditions of oxygen saturation no after-effects could be obtained. The experiment illustrates another finding that I have noted, namely, that it is in early stages of anoxemia that the auricles exhibit a tendency to develop after-effects, generally the late effect of anoxemia is to inhibit the onset of fibrillation.

TABLE 5
Dog 35 Vagus nerves tied, no atropine used

Oxygen saturation of blood	Number of faradic stimulations	Duration of after-effect	
		15 to 59 seconds	1 minute or longer
<i>per cent</i>			
100	50 (0)*	0	0
73	8 (0)	1	1
100	5 (3)	0	0
73	5 (5)	2	3
100	4 (4)	0	2
65†	7 (7)	0	0

* The figures in the parentheses indicate the number of times vagal stimulation was used together with faradic stimulation

† Approximately

The findings in these and other experiments afford an explanation for the one instance (experiment 22), in which there was a greater tendency for fibrillation to appear when the blood was normally saturated with oxygen than when anoxemia was present. In this case, a moderate anoxemia (80 per cent saturation) had been present for slightly more than two hours.⁴ A high oxygen mixture was then given, and faradic stimulation produced three short and two long after-effects in thirty trials. On lowering the oxygen saturation of the blood to 80 per cent, fifty trials caused only one long after-effect. The reason for this exceptional result is undoubtedly due to the fact that *functionally* the auricles during the period when the blood was normally saturated, were in a state of mild anoxemia due to incom-

⁴ During this time other observations were being made

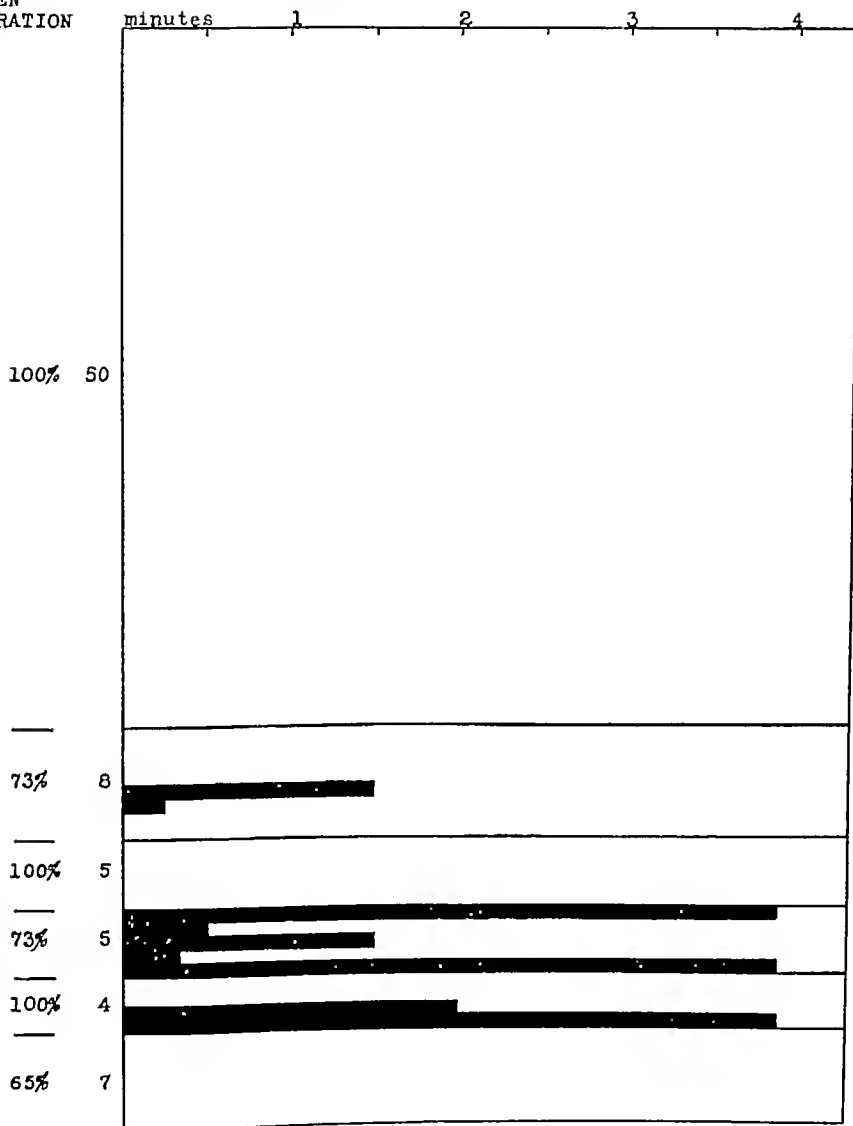


FIG 2 EXPERIMENT 35 THE RESPONSE OF THE AURICLES TO FARADIC STIMULATION, VAGAL TONE LARGELY ELIMINATED, DURING PERIODS OF NORMAL AND LOW OXYGEN SATURATION OF THE BLOOD

On the left hand side of the chart are given the percentages of oxygen saturation of the arterial blood and the number of faradic stimulations during the different periods. The width of a single black bar represents a single after-effect, its length showing the duration of the after-effect. The stippled areas mean that the after-effect in these instances lasted longer than the chart indicates. Note the increased tendency for fibrillation to develop during the early periods of anoxemia (second and fourth blocks of the chart), the persistence of this tendency when normal saturation is restored after a preceding period of anoxemia (fifth block), and the failure of fibrillation to appear in the late stages of anoxemia (lowest block).

plete recovery from the previously existent anoxemia of long duration. When the oxygen saturation of the blood was then reduced, the late effect of anoxemia was seen, that is to say, auricular fibrillation was hindered.

There were three experiments in which no difference could be detected in the response to faradic stimulation, either with normal or low oxygen mixtures. In two of these, no after-effect could be produced. It must be recalled that atropine places a serious obstacle in the way of producing auricular fibrillation by any method. Consequently, it does not necessarily follow that anoxemia was without effect in these two cases. It may well have been that the anoxemia could not overcome the antagonistic influence of atropine.

One may summarize the foregoing data by stating that anoxemia predisposes the auricles to fibrillation, even when vagal tone is completely removed. Recovery of the auricles from a previous period of anoxemia is not immediate, and after-effects may be observed in incompletely recovered auricles when the blood is normally saturated with oxygen. Whereas previously, under the same conditions of oxygen saturation of the blood, after-effects may have been absent. Anoxemia exerts that influence on the auricles which predisposes to fibrillation, relatively early, and the late effect of anoxemia is to inhibit the development of auricular fibrillation.

The refractory period of the auricle. In seven experiments, determinations of the refractory period of the auricles were made during periods of normal and diminished oxygen saturation of the blood. It was found that anoxemia first decreased the refractory period, but later increased it, bringing on various degrees of intra-auricular block. In table 6 are the data of an experiment illustrating the diminution of the refractory period by anoxemia.

In this table are given the figures for the latest ineffective⁵ and the earliest effective shocks, the calculated refractory period being

⁵ In this and following tables, the "latest ineffective stimulus" does not actually represent the longest interval between rhythmic and interrupting shocks at which a response occurs. It represents the interval below which all other interrupting stimuli are ineffective. A certain amount of overlapping of effective and ineffective shocks is prone to occur when the auricles are atropinized, particularly at the higher rates of driving.

TABLE 6

Dog 126

Time	Oxygen saturation 96 per cent		Oxygen saturation 50 per cent started 11 30		Oxygen saturation 96 per cent started 12 03	
	12 05	11 20	11 42	11 47	12.30	12 40
Auricular rate per minute	216	273	223	273	222	276
Latest ineffective stimulus	0 092	0 081	0 083	0 073	0 095	0 087
Earliest effective stimulus	0 092	0 082	0 087	0 077	0 103	0 087
Calculated refractory period	0 092	0 0815	0 085	0 075	0 099	0 087

TABLE 7

Dog 30 Auricular rate 295 per minute

Time	Oxygen saturation	Latest ineffective stimulus	Earliest effective stimulus	Calculated refractory period
	<i>per cent</i>			
12 11	100	0 090	0 097	0 0935
12 14	73			
12 38	73	0 076	0 078	0 077
12 41*	†	0 095	0 101	0 098
12 46	†	0 106	‡	>0 106

* Two minutes after appearance of ventricular fibrillation

† No figures are given for the percentage of arterial oxygen saturation as this must have decreased very rapidly during ventricular fibrillation

‡ There were no instances in which an interrupting shock was preceded by two effective rhythmic shocks, a necessary precaution in the determination of the refractory period (Lewis, Drury and Bulger, 1921) Consequently, no determination was made for the earliest effective stimulus at this time

TABLE 8

Dog 27 Auricular rate 221 Refractory period at rate of 214 was 0 1095 with normal arterial oxygen saturation

Time	Oxygen saturation 45 per cent Started 1 35		
	Latest ineffective stimulus	Earliest effective stimulus	Calculated refractory period
1 57	0 093	0 093	0 093
2 04	0 082	0 090	0 086
2 10	0 087	0 091	0 089
2 19	0 098	0 098	0 098
2 32	0 092	0 100	0 096
2 40	Intra-auricular block	—	—

taken as the average of the two figures. In this experiment, shortly after producing a severe anoxemia, the refractory period decreased slightly but definitely, returning to figures above those originally obtained, when the normal oxygen saturation of the blood was restored.

In tables 7 and 8 are recorded two further experiments in which anoxemia was continued until the animals died. In both there is seen a fall, then a rise in the refractory period during anoxemia. In each of these experiments the refractory period increased only when the animal was in very poor condition.

TABLE 9

Time	Oxygen saturation of blood	Rate of auricle
	<i>per cent</i>	<i>per minute</i>
12 23	100	659
12 25	50	
12 36	50	621
12 40	50	608
12 45	50	612
12 50	50	614
12 55	50	590
1 00	50	549
1 05	50	480
1 05 30	50	465
1 06	50	440
1 07	50	Normal mechanism*

* The animal died a few minutes after the reversion to normal mechanism.

In two of the four remaining experiments, there was a decrease in the refractory period, which occurred a short time after a moderate anoxemia was started. In the other two experiments only the stage of increase in the refractory period was obtained, in one the anoxemia was severe (47 per cent arterial oxygen saturation), in the other, the anoxemia was mild (oxygen saturation, 89 per cent), but of relatively long duration, lasting almost two hours.

The data on the effect of anoxemia on the refractory period of the auricle are admittedly insufficient to give a complete picture since the rhythmic rates were relatively low in the determinations which I made. They are presented because they are consistent, in a general way, with the other observations on the effect of anoxemia.

Effect of anoxemia on auricular fibrillation already present On several occasions the effect of anoxemia on the rate of a circus movement was noted. Experiment 21 (table 9) is illustrative of the others.

Eleven minutes after the onset of anoxemia, there was a slight decrease in the rate of fibrillation, from 659 to 640 per minute, the level then remained about the same for fourteen minutes more. The rate then fell again, gradually at first, but very rapidly before the spontaneous reversion to the normal mechanism took place.

In one animal (experiment 35, table 10) flutter was produced shortly after anoxemia was induced. The rate of the auricle diminished

TABLE 10

Dog 35 Vagus nerves tied, atropine not used Auricular flutter started at 12 05

Time	Oxygen saturation of blood	Auricular rate
	<i>per cent</i>	<i>per minute</i>
12 06	73	600
12 08	73	590
12 11	73	582
12 18	73	580
12 29	73	582
12 30	100	
12 31	100	583
12 33	100	588
12 40	100	588
12 46	100	588
12 50	100	587
12 50 15	100	Normal mechanism

gradually from 600 to 582 per minute within five minutes and the rate remained constant for eighteen minutes. A high oxygen mixture was then given. The rate rose to 588 within three minutes, remained at this level for seventeen minutes and almost immediately after the last record was taken (the rate of which was 587) the normal mechanism was restored.

DISCUSSION

The results which were obtained with faradic stimulation during anoxemia indicate definitely that anoxemia predisposes the auricles to fibrillation, and this effect is the more striking in that it may overcome the powerfully antagonistic influence of atropine. I have

purposefully stated that anoxemia *predisposes* the auricles to fibrillation in none of these experiments has auricular fibrillation appeared spontaneously. Apparently some other factor besides anoxemia is necessary to initiate this arrhythmia. In these observations, using the normal heart of the dog, this additional factor was the rapid stimulation of the auricles by a faradic current. What this second factor may be in the various clinical conditions in which anoxemia seems to play a rôle in causing transient auricular fibrillation, it is difficult to determine. Undoubtedly this influence differs in the individual case, although fundamentally its effect is probably always the same.

The development of a circus movement depends essentially upon shortening of the refractory period and lengthening of the rate of conduction, these occurring in such a fashion that the slowly travelling wave of excitation is always able to enter responsive muscular tissue, the refractory state having passed off. When the two factors of primary importance, namely refractory period and rate of conduction, are altered in such a way that they are mutually antagonistic the outcome depends on the preponderating effect of one or the other of the actions. Thus, at high rates of stimulation atropine slows the rate of transmission of the excitation wave, but the opposite influence of the increased refractory period practically always predominates and auricular fibrillation is hindered. Conversely, vagal stimulation increases the rate of transmission through the auricle at high rates of stimulation, but the absolute refractory period is shortened at the same time, and usually the appearance of auricular fibrillation is favored.

At first sight it would appear that the effect of anoxemia on the refractory period explains the type of response of the auricles to faradic stimulation in the early and late stages of anoxemia. It is in the early stage that the auricles show a predisposition to the development of fibrillation, and it is at this time that the refractory period is shortened. Likewise, the failure of fibrillation to appear (usually) in the late stages of anoxemia is apparently explained by the increase in the refractory period. Unfortunately I have been unable to make determinations of the rate of transmission of the excitation wave through the auricular muscle, nor have I obtained satisfactory ob-

servations on the effect of anoxemia on the refractory period at high rates of stimulation I have pointed out elsewhere (Resnik, 1925a) that at relatively low rates of stimulation, auriculo-ventricular conduction may be quickened by anoxemia, whereas higher rates may cause conduction defects to appear. It is quite possible that the same events may take place in the refractory period of the auricles. Indeed, from the response to anoxemia of a circus movement already established (tables 9 and 10) it seems certain that the simple shortening of the refractory period cannot entirely account for the development of auricular fibrillation in the early stages of anoxemia, for if it were the case, the rate of the circulating rhythm should increase, for a short time at least, on changing from normal to low oxygen saturation of the blood. The shortening of the refractory period, if it were the only change to occur, should then permit the excitation wave to take a shorter path, this in turn allowing the excitation wave to traverse its course a greater number of times per minute. However, I have found that soon after diminishing the percentage of oxygen saturation of the blood, the rate of the circus movement decreased. Without a knowledge of the actual changes which occur in the refractory period and the rate of conduction when the auricles beat rapidly during anoxemia, it is useless to pursue the discussion. With the present data, one can state that, whatever may be the absolute alterations of refractory period and conduction, *relatively* the refractory period is decreased and conduction slowed, in the early stages of anoxemia, so that a circus movement is permitted to develop, in the late stages of anoxemia the reverse is true.⁶

SUMMARY AND CONCLUSIONS

1 The sino-auricular node is highly sensitive to anoxemia. There is at first a brief period during which the rate of impulse formation is

⁶ No reference has been made as to the ultimate cause by which slowing of the rate of transmission of the excitation wave through the auricles may be brought about. Drury (1925) describing work by himself and Andrus has presented evidence to show that injury of the muscular tissue by various means causes the phenomenon known as "conduction with decrement" to appear. Whether this is true of conduction defects in the auricles due to anoxemia (and it is highly probable that they exist), I am unable to state. Since this paper was written, Drury's article has appeared in *Heart*, 1925, xii, 143.

accelerated, followed rapidly by progressive slowing of the intrinsic rate of the heart

2 Relatively low rates of rhythmic stimulation, anoxemia first reduces, then increases the absolute refractory period of the auricles

3 Anoxemia diminishes the rate at which a circus movement travels throughout the auricle

4 When vagal tone is removed, and the auricles are stimulated by a faradic current, the early effect of anoxemia is to predispose the auricles to fibrillation, the late effect is to inhibit the development of fibrillation. The mechanism by which anoxemia produces these effects is discussed

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SIMULTANEOUS RESPIRATORY EXCHANGE AND BLOOD SUGAR TIME CURVES OBTAINED IN DIABETIC AND NON DIABETIC INDIVIDUALS FOLLOWING INGESTION OF GLUCOSE

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(Received for publication July 1, 1925)

INTRODUCTION

Hyperglycaemia and glycosuria result when the combined rates of oxidation and storage do not keep pace with the rate of absorption of glucose from the alimentary canal or its formation in the body. Carbohydrate tolerance is therefore usually estimated by the study of glycaemia and glycosuria. Another generally recognized, and probably more efficient method, is the study of the respiratory exchange. These procedures have, from time to time, depending upon the circumstances in any given case, been employed independently. It would appear, however, that a study combining both methods would be the most satisfactory.

Even in normal individuals simultaneous blood and respiratory metabolism data obtained following the ingestion of glucose are very scanty. In isolated instances in studying carbohydrate metabolism in diseases other than diabetes, such data have been obtained for comparative purposes. Of special note are those of Sanger and Hun (1) in their work on hyperthyroidism and Linder, Hiller and Van Slyke (2) on nephritis. Finley and Rabinowitch (3) and more recently Ladd and Richardson (4) applied this method in a study of renal glycosuria.

The object of this paper is to record simultaneous respiratory exchange and blood sugar time curves obtained in five cases of renal glycosuria, eleven cases of diabetes of different severity, and two cases of pituitary disease with glycosuria.

For the discussion of results it is necessary first to consider briefly the normal values obtained with the individual procedures. The literature of each method has frequently been reviewed and requires no repetition.

BLOOD SUGAR

In a comprehensive study, including a statistical analysis of 4000 individual determinations in 971 cases, Gray (5) presented the views now most generally accepted. The values of the mean and the mode (the most typical results) of the series agreed very closely. The blood sugar time curve is characteristic. The fasting blood sugar is about 0.09. Following the administration of glucose, the maximum increment is noted within 30 minutes, and the value is below 0.180 per cent, the average peak being 0.140 per cent. The rate of decline is at first more rapid, then slower. Within 3 hours the blood sugar concentration is equal to or below that of the fasting period.

In only ten of 431 cases was the fasting level above 0.12 per cent. Eight of these were 0.130 per cent. In one case it was 0.15 and in another 0.160 per cent. These data are of particular interest since, in the majority of cases (300 out of 479), the dose of sugar administered by the various workers was that now generally employed, namely, 70 to 100 grams. In the majority of the 300 cases abnormal glycosuresis did not occur. It was, however, found in a sufficient number, namely, 129, to emphasize the difficulty of the interpretation of this phenomenon.

The majority of observers are also in agreement as to the relation between the type of the curve and the dose of sugar given. With as little as 5 grams an appreciable increase was noted. The highest peak with the smallest dose was as great as the average peak with the larger dose. With doses exceeding 25 grams the peak was not affected. Increasing the dose merely prolonged the time necessary for the blood sugar to return to its original fasting level. The difference between the effects of the starches and glucose was too small to determine any special preference.

In diabetics (excluding hypertension, nephritis, hepatic disorders, hyperthyroidism, hyperpituitarism, etc.) the mean values corresponds to those now generally recognized. Following the administration of

glucose the rate of increase of the blood sugar percentage is slower and the peak occurs later (at the end of the first hour) The rate of return to the original level varies with the severity of the disease as indicated by the degree of hyperglycaemia in the post-absorptive state

In renal glycosuria, in the absence of nephritis, the mean levels of the fasting blood sugar, the rate and the character of the curve of decline are essentially the same as those found in normal individuals

RESPIRATORY EXCHANGE

The average respiratory quotient during the fasting period is 0.82 (0.83 for males and 0.81 for females) (6) After the administration of 100 grams of glucose an increase may be noted within 7 minutes (7) and rarely fails to occur within 30 minutes The average maximum increase is about 0.12 (8) In some instances a slight decrease may be noted In conditions which deplete glycogen, such as exercise, starvation, etc., no increase or an actual decrease may be noted for a time (7, 8, 9) Benedict and Carpenter (8) found a fairly consistent picture in the ten cases observed The lowest, highest and average peaks of carbon dioxide production were respectively 12, 35 and 25 per cent above the basal period, of oxygen consumption, 3, 22, and 12 per cent, of the heat production 6, 24 and 14, and of the respiratory quotients, 0.07, 0.17 and 0.12 per cent The average time of appearance of the maximum effects was within 2 hours

Excluding the technical difficulties, psychic and other factors which may yield artificial respiratory quotients—common to both normal and diabetic subjects—and assuming that the quotients obtained approximate true metabolic respiratory quotients, there are still many factors which influence their value in the fasting state in diabetes and make difficult their interpretation Rate of metabolism composition of the diet of the previous day the oxidation of acetone bodies non-utilization of carbohydrates, incomplete oxidation of fats and decreased urea formation are some of these factors The numerous vagaries have been discussed by both Joslin and DuBois in their recent monographs (10) It is therefore unnecessary to repeat these here

Following the administration of glucose to a diabetic in the absence of insulin, three types of response may be noted (a) no increase (b)

an increase, and in some cases approximating the response in normal individuals, and (c) an initial decrease followed by an increase. All three types of response were found during this investigation. These will be discussed separately, and then correlated with the clinical picture and blood sugar curves. The curve of the heat production may or may not parallel the curve of the respiratory quotient.

METHODS

Classification of cases

The diagnosis of all the cases of renal glycosuria was based upon the generally accepted clinical and laboratory standards, namely, (a) absence of hyperglycaemia, (b) no uniform relation between intake and excretion of sugar, (c) excretion of sugar slight, (d) normal blood sugar time curves following administration of glucose, and (e) absence of other clinical signs and symptoms suggestive of the common type of diabetes. The important clinical index pointed out by Joslin, namely, the absence of diabetes following many years of observation, could for obvious reasons not be applied.

The classification of the diabetics into very mild, mild (incipient), moderate and severe, must, of necessity, be approximate. Joslin, though not strictly adhering to carbohydrate tolerance as a guide, does employ it. The individuals with a tolerance of less than 10 grams of glucose are regarded as severe, between 10 and 50 grams as moderate, and with a tolerance of 50 grams or over as mild. A classification based upon the use of insulin is suggested. According to Von Noorden's classification, those individuals are regarded as severe, who, in spite of a prolonged and extremely low carbohydrate diet, fail to become sugar free. Woodyatt¹ suggests that the only rational method of measuring the degree of diabetes is to measure the velocity at which glucose can be utilized, that is, the number of grams utilized per unit of time such as 24 hours. The utilization of glucose equals the supply minus the excretion. The classification employed in this hospital, and for this investigation, combines that of Joslin and Von Noorden, with certain modifications. It is based upon (a) glycosuria, (b) acc-

¹ Personal communication

tonuria, in the absence of any previously restricted diet, (c) clinical signs and symptoms, (d) response to a "green day" diet, and (e) composition of the final diet

1 Very mild The glycosuria is transient or post prandial only, and disappears readily with moderate change in diet, which includes eliminating the sugar in tea or coffee avoiding pastries and puddings, but allowing one ounce of bread at each meal. Otherwise no particular restriction is made as to meats, or vegetables. The glycosuria is discovered by routine examination, and there are no other signs or symptoms of diabetes.

2 Mild cases The glycosuria is present throughout the day, but may be made to disappear in one or two days, following a "green day" diet, consisting of 15 grams of carbohydrates, 20 grams of fat and 25 grams of protein. These patients following a "ladder diet," eventually tolerate as much as 150 grams carbohydrates. Clinical signs and symptoms may or may not be present. In the absence of a previous restriction of diet the urine contains no acetone bodies.

3 Moderate The glycosuria is persistent throughout the day. Acetone bodies are found in the urine in the absence of previous restriction in diet. Clinical signs and symptoms are more pronounced. The urine becomes sugar free following 3 to 4 "green" days. The final diet contains at least 50 grams of carbohydrates, 150 grams of fat, and 50 grams of protein.

No insulin is required to maintain the average weight for the height.

4 Severe The patient does not become sugar free after 4 "green days." Acetone bodies in the urine are marked. The clinical symptoms are still more pronounced. Glycosuria persists throughout the "ladder" period, while the diet is approaching the basal requirements. Insulin is necessary with the final diet, to maintain the average weight for the height of the individual.

LABORATORY TECHNIQUE

All tests were performed in the morning. The respiratory quotient and basal metabolic rate were first obtained in the post-absorptive state, and after half an hour of complete muscular rest. Blood and urine specimens were then taken. The dose of glucose (dissolved in 150 cc. of water and flavoured with lemon juice) as indicated in the tables, was then given. The respiration experiments were of 10 minutes duration each, and commenced at such a time so that each period ended at the time recorded in any given case, namely 30 minutes, 60 minutes, 1 hour, 2 hours etc. following the ingestion of glucose. Blood and urine specimen were obtained at the end of each respiratory experiment. The Tissot gasometer and Haldane gas analysis apparatus were used. All gas analyses were made in dupli-

cate by the same individual (E V B) All blood and urine estimations were made by the same individual (A B F) For the blood sugars the modified Myers-Benedict method was used Urine sugars were estimated by the Benedict-Osterberg procedure after suitably diluting the specimens

CALCULATIONS

In the cases of renal glycosuria, very mild—mild, and in some moderate cases of diabetes, a constant protein metabolism was assumed (15 per cent of the total) On this basis the non-protein respiratory quotients were calculated In some moderate cases and in the severe type of diabetes associated with abnormal metabolism, nitrogen determinations were made by the Kjeldahl method, and thus non-protein respiratory quotients were obtained From the non-protein respiratory quotient and the metabolic rate the calculations for the utilization of the carbohydrate were made in the usual manner The non-protein calories, assumed to be derived from the carbohydrates and fats, were apportioned according to the Zuntz-Schumberg Tables as modified by Williams, Riche and Lusk (11) The results are recorded in the accompanying tables No alterations were made in the calculations where the respiratory quotients were greater than 1.0 or less than 0.70 In the former case, carbohydrate, and in the latter, fat oxidation, was assumed to be the only factor influencing the respiratory quotients

DISCUSSION

The detailed data obtained from the cases of renal glycosuria are recorded in table 1 The average increment in carbon dioxide production was 26.4 per cent, in oxygen consumption 9.6 per cent, in heat production 13 per cent, and in the respiratory quotient 0.11 These data approximate very closely those obtained by Benedict and Carpenter on normal individuals The average amount of glucose oxidized per hour for the first 2 hours was 11.99 grams This also approximates very closely that recorded by Finley and Rabinowitch in one case of renal glycosuria (3) and other unpublished data obtained in normal individuals in this laboratory The above data therefore, further strengthen the view that the glycosuria in this condition results

from an undue permeability of the kidneys, the functions of oxidation and storage, unlike these in true diabetes, remaining unimpaired. In

TABLE 1

Simultaneous respiratory exchange and blood sugar time curves in cases of renal glycosuria

Time	O ₂ per hour	CO ₂ per hour	Total R Q	Nitrogen per hour	Non protein R Q	Calories per hour	Carbohydrate per hour	Blood sugar	Remarks
minutes	liters	liters		grams			grams	per cent	
Basal	16 58	11 87	0 716	0 474	0 698	77 8	0	0 119	No 1 Renal glycosuria, 100 grams glucose
30								0 166	
60	16 76	13 64	0 814	0 492	0 816	80 7	6 47	0 111	
120	16 04	13 47	0 84	0 474	0 847	77 8	8 64	0 125	
180								0 087	
Basal	11 64	8 95	0 769	0 338	0 761	55 5	2 31	0 094	No 2 Renal glycosuria (pregnancy), 100 grams glucose
30								0 144	
60	12 36	11 33	0 917	0 401	0 943	65 2	11 74	0 102	
120	13 24	12 03	0 909	0 398	0 931	65 4	11 29	0 119	
180								0 068	
Basal	12 48	8 78	0 704	0 357	0 683	58 5	0	0 103	No 3 Renal glycosuria, 100 grams glucose
30								0 108	
60	13 91	12 15	0 874	0 414	0 887	68 0	9 64	0 161	
120	11 74	10 21	0 870	0 350	0 883	57 4	7 70	0 120	
180								0 096	
Basal	16 50	11 74	0 712	0 473	0 692	77 4	0	0 074	No 4 Renal glycosuria, 100 grams glucose
30								0 154	
60	18 85	14 89	0 790	0 550	0 787	90 3	5 85	0 159	
120	18 86	15 40	0 817	0 554	0 821	91 0	8 00	0 099	
180								0 077	
Basal	17 84	13 05	0 732	0 512	0 716	84 1	0 266	0 077	No 5 Renal glycosuria 100 grams glucose
30								0 123	
60	17 45	12 37	0 709	0 550	0 686	91 8	0	0 090	
120	18 81	14 82	0 788	0 549	0 784	90 1	5 14	0 100	
180								0 085	

one case (no 5) there was noted a fall in the total respiratory quotient as late as 1 hour after the ingestion of glucose and all the non-protein respiratory quotients were below 0 70. It is thus assumed that no

carbohydrate was being oxidized during the period of investigation. This case is of particular interest. The fall in the respiratory quotient and the absence of oxidation of glucose may be attributed to the fact that the patient was in a state of carbohydrate starvation at the time of the test. Because glycosuria was found, the patient was apprehensive of his condition, and lived on a low carbohydrate diet for weeks prior to the test. The importance of recognizing this phenomenon in the interpretation of the results of blood sugar time curves in diabetes will be discussed later.

In table 2 are recorded the detailed data from the different types of cases of diabetes, very mild, mild, moderate and severe. Since the data of the individual subjects are so at variance with one another and the cases differed clinically, a study of average results would appear to be of little value. It will be noted that in all the cases the rate of utilization of glucose (grams in 2 hours), was much below the average found for normal individuals. There was, however, no perfect correlation between the rate of utilization and the severity of disease. An explanation of this is suggested in the following discussion.

In the majority of cases recorded in the literature no rise in the respiratory quotient is noted following the ingestion of glucose. This is naturally attributed to the inability of the diabetic to tolerate carbohydrates. It might be pointed out that no rise in the respiratory quotient may indicate defective oxidation. It does not, however, necessarily follow that there is also defective storage. Tolerance for carbohydrates includes both oxidation and storage. In the cases recorded here three different types of reactions to glucose ingestion, manifested in the respiratory quotient, are noted, namely, (a) no response, (b) an increment, and (c) an initial decrement followed by an increment.

All the respiratory quotient curves and their corresponding blood sugar curves are recorded in table 3. This is done with the object of correlation. Unexpected results were found. It will be noted that as late as at the end of one hour in the very mild (no. 6), in one mild (no. 7), and in two moderate (nos. 9 and 10) cases, there was an initial decrement in the respiratory quotient. On the other hand, in two of the three severe cases (nos. 14 and 15), a definite increment was noted. In both of the latter cases the heat production had also increased.

TABLE 2

Simultaneous respiratory exchange and blood sugar time curves in cases of diabetes mellitus

[illegible]

TABLE 2—*Continued*

Time	O ₂ per hour	CO ₂ per hour	Total R Q	Nitrogen per hour	Non protein R Q	Calories per hour	Carbohydrate per hour	Blood sugar	Remarks
<i>minutes</i>	<i>liters</i>	<i>liters</i>		<i>grams</i>			<i>grams</i>	<i>percent</i>	
Basal	10 60	7 36	0 696	0 302	0 672	49 7	0	0 175	No 12 Moderate type, 50 grams glucose
30								0 263	
60	10 61	7 70	0 726	0 304	0 708	49 9	0 158	0 312	
120	11 09	8 38	0 756	0 320	0 745	52 6	1 11	0 357	
180								0 277	
Basal	14 84	10 90	0 735	0 426	0 723	70 0	0 76	0 104	No 13 Moderate type, 50 grams glucose
30								0 212	
60	17 23	14 88	0 864	0 512	0 878	84 0	11 27	0 147	
120	15 83	13 47	0 851	0 469	0 860	77 0	10 47	0 076	
180								0 05?	
Basal	11 91	8 53	0 716	0 341	0 698	55 9	0	0 200	No 14 Severe type 25 grams glucose, urine—sugar and diacetic acid at time of test
30								0 316	
60	12 50	9 53	0 762	0 362	0 753	59 4	2 01	0 324	
120	12 40	9 29	0 749	0 317	0 738	58 8	1 57	0 320	
180								0 323	
Basal	11 03	7 62	0 691	0 315	0 667	51 7	0	0 204	No 15 Severe type, 25 grams glucose, urine—sugar and diacetic acid at time of test
30								0 256	
60	11 90	8 96	0 753	0 343	0 742	56 4	1 47	0 284	
120	12 28	9 00	0 733	0 360	0 718	57 9	0 62	0 276	
180								0 278	
Basal	12 90	9 25	0 717	0 369	0 708	60 7	0	0 210	No 16 25 grams glucose, severe type, urine—sugar and diacetic acid at time of test
30								0 246	
60	13 06	9 40	0 720	0 374	0 702	61 4	0	0 278	
120	13 18	9 38	0 712	0 377	0 693	61 8	0	0 302	
180								0 300	

The subjects with severe diabetes were apparently oxidizing glucose, but in spite of this the blood sugars were increasing. It appears reasonable therefore, that the hyperglycaemia was due chiefly to defective storage. McCann and Hannon (11) in a study of the respiratory exchange alone, obtained similar results, and offered an explanation of this phenomenon. The data obtained in the cases recorded here, by a simultaneous study of the respiratory exchange and blood sugar, strengthen that view.

There appear to be two different types of diabetics, namely, those who can store more readily than oxidize and those who can oxidize more readily than they can store. To the former group appears to belong the less severe type of the disease. It will be noted that the fall in the respiratory quotient was noted in the very mild, mild and moderate cases. The level to which the respiratory quotient may fall is striking. That such low quotients are no index of severity of the disease is further suggested from the fact that Benedict and Carpenter (8)

TABLE 3
Summary of respiratory quotient and blood sugar data

Case number	Respiratory quotients			Blood sugar per cent					Remarks
	Basal	60	120	Basal	30	60	120	180	
1	0.716	0.814	0.840	0.119	0.166	0.111	0.125	0.087	Renal glycosuria
2	0.769	0.917	0.909	0.094	0.144	0.102	0.119	0.068	
3	0.704	0.874	0.870	0.103	0.108	0.161	0.120	0.096	
4	0.712	0.790	0.817	0.074	0.154	0.159	0.099	0.077	Very mild diabetic
5	0.732	0.709	0.788	0.077	0.123	0.090	0.100	0.085	
6	0.732	0.684	0.726	0.125	0.133	0.137	0.105	0.100	
7	0.808	0.875	0.864	0.127	0.176	0.238	0.312	0.344	Mild diabetic
8	0.689	0.676	0.727	0.116	0.178	0.196	0.256	0.250	
9	0.747	0.674	0.732	0.196	0.228	0.254	0.252	0.250	
10	0.784	0.724	0.736	0.118	0.184	0.207	0.156	0.137	Moderate
11	0.661	0.728	0.695	0.164	0.250	0.248	(?)	0.232	
12	0.696	0.725	0.756	0.175	0.263	0.312	0.357	0.277	
13	0.735	0.864	0.851	0.104	0.212	0.147	0.076	(?)	Severe
14	0.716	0.762	0.749	0.200	0.316	0.324	0.320	0.323	
15	0.691	0.753	0.733	0.204	0.256	0.284	0.276	0.278	
16	0.717	0.720	0.712	0.210	0.264	0.278	0.302	0.300	Pituitary disease, glycosuria
17	0.855	0.941	1.01	0.103	0.133	0.158	0.107	0.104	
18	0.727	0.828	0.767	0.100	0.088	0.111	0.083	0.095	

found such a decrease (0.70 to 0.67) in a normal individual (B. M. K.) following the ingestion of 100 grams dextrose, and McCann (13) noted such a decrease (0.72 to 0.68) in a normal individual after protein ingestion.

On the whole, no perfect correlation is noted between the respiratory quotient and blood sugar curves. This is to be expected. The type of respiratory quotient curve obtained depends upon the character of the reaction to glucose ingestion, namely, storage or oxidation.

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THE EFFECT OF CAFFEIN SODIO-BENZOATE THEOBROMIN SODIO-SALICYLATE THEOPHYLLIN AND EUPHYLLIN ON THE CORONARY FLOW AND CARDIAC ACTION OF THE RABBIT¹

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(Received for publication July 13, 1925)

INTRODUCTION

The results of the investigation of the action of caffeine, theobromin and theophyllin on the coronary arteries vary considerably with the different observers. Hedbom (1) employing the Langendorf method, produced an increase in coronary flow which, in some instances was said to be great with caffeine in concentrations of 1-20 000. Loeb (2) on the other hand observed very little increase in the coronary output following the administration of caffeine whereas theobromin greatly augmented the rate of perfusion. Heathcote (3) working with solutions of caffeine, theobromin and theophyllin in concentrations ranging from 1-2 000 to 1-40 000 obtained a marked increase in rate of coronary flow with the higher concentrations, whereas in dilutions of 1-20 000 and 1-40 000 caffeine produced no change, theobromin very slight and theophyllin 20 to 30 per cent increase. Sakai and Saneyoshi (4), from experiments on the intact heart of the cat in which they employed the Morovitz-Zahn cannula concluded that the increase in the coronary output which they observed following the injection of caffeine sodio-benzoate could satisfactorily be explained on the basis of the elevation in blood pressure. These investigators reported a slight decrease in the coronary flow with small doses of theobromin sodio-salicylate, whereas, when large doses of the drug were

¹ Read before The American Society for Clinical Investigation at Washington, D. C., May 4, 1925

introduced, there was a drop in blood pressure associated with a marked augmentation of the flow through the coronary arteries

Guggenheimer and Sassa (5) have more recently checked the action of caffeine, theophyllin and euphyllin on the coronary arteries of the isolated heart of the cat. Caffeine in a dilution of 1-25,000 increased the rate of coronary flow 41 per cent. Theophyllin and euphyllin, in similar concentrations, produced a 40 and 80 per cent increase, respectively. They attributed the change in the rate of the flow of the perfusate after caffeine and theophyllin to the acceleration of cardiac rate and pointed out that euphyllin did not elevate heart rate more than 10 per cent. The latter drug, which is a combination—80 per cent theophyllin and 20 per cent ethylenediamine, was recommended therapeutically in coronary artery disease because of its marked dilating action.

The object of the present investigation was to substantiate, if possible, the claims made by Guggenheimer and Sassa (5) for euphyllin and to compare the action of this drug with that of caffeine sodiumbenzoate, theobromin sodiumsalicylate and theophyllin in experiments in which the cardiac rate was controlled.

METHODS

The isolated heart of the rabbit was employed. The rabbit was killed in the usual manner, the chest quickly opened, the heart removed and placed in a basin containing Locke-Ringer solution. The blood was carefully washed from the left ventricular cavity in order that it might not coagulate, and bits of coagulum later be thrown into the coronary circulation. A cannula was then introduced into the aorta, and the heart perfused at a pressure of 55 mm Hg with a Locke-Ringer solution saturated with oxygen. The temperature of the perfusate was maintained at a constant level of 100° by a series of coils passing through a water bath. The control temperature was registered by a thermometer introduced into the cannula connecting directly with the aorta.

The rate of the coronary flow was determined by allowing the fluid which escaped from the heart to pass through a small funnel into a tipping bucket, the emptying of which was automatically registered on a kymograph. It is recognized that this method of measuring the coronary flow does not take into consideration the possibility of leakage through the aortic valves. This feature was, however, carefully investigated in a series of experiments² and found to be negligible.

² In a series of experiments a cannula was placed in the pulmonary artery. The return of the perfusate to the right heart was thus separated from any that might

The cardiac rate and amplitude were registered by means of a lever connected to the apex. In the records, the upstroke represents the cardiac contraction and the down stroke relaxation.

The concentration of the drug employed varied from 1-25,000 to 1-50,000² which was estimated to be within the range of the concentration obtained in the blood in man following the administration of a therapeutic dose. The drug was not administered until the cardiac rate, amplitude of contraction and the coronary flow assumed a uniform level. In each instance the condition of the heart remained satisfactory throughout the experiment. The results were discarded in those experiments in which the heart failed to return to a satisfactory condition from the standpoint of amplitude of contraction and rate of coronary flow following the discontinuation of the drug.

In those instances in which there was an increase in heart rate from the action of the drug, the cause of the increased coronary flow was verified by experiments in which a uniform cardiac rate was maintained by rhythmically induced break shocks. The rhythmical stimulation of the heart was produced by an oscillating contact maker, employing the principle of the tuning fork, which was connected with an ordinary induction coil. The rate of stimulation used was always well above that produced by the action of the drug on the heart with normal mechanism. In most instances the rate employed was 224 per minute.

RESULTS

Caffein sodio-benzoate in concentration of 1-25,000 solution had little or no effect on the cardiac rate, amplitude of contraction and coronary flow (fig 1).

Theobromin sodio-salicylate in dilution of 1-25,000 in most instances produced no change, or even decreased the coronary flow (fig 2). In one instance in which 1-12,500 concentration was employed there was 17.6 per cent reduction in the rate of perfusion. When, however, the concentration of 1-50,000 was used, the rate of flow was slightly accelerated. In two instances the percentage of increase was 25 and 28 per cent, respectively. Theobromin sodio-salicylate in either of the above concentrations apparently had no

leak past the aortic valves or return directly into the left ventricle from the coronary circulation. The same feature was investigated by ligating the pulmonary veins and inserting a cannula in the tip of the left auricle. In both instances the amount of perfusate which passed from the left heart was negligible.

² A dilution of 1-50,000 was estimated to correspond to a dose in man of 1.5-2.5 grains, depending on the size of the individual, a dilution of 1-25,000 to a dose 3-5 grains and a dilution of 1-12,500 to a dose of 6-10 grains.

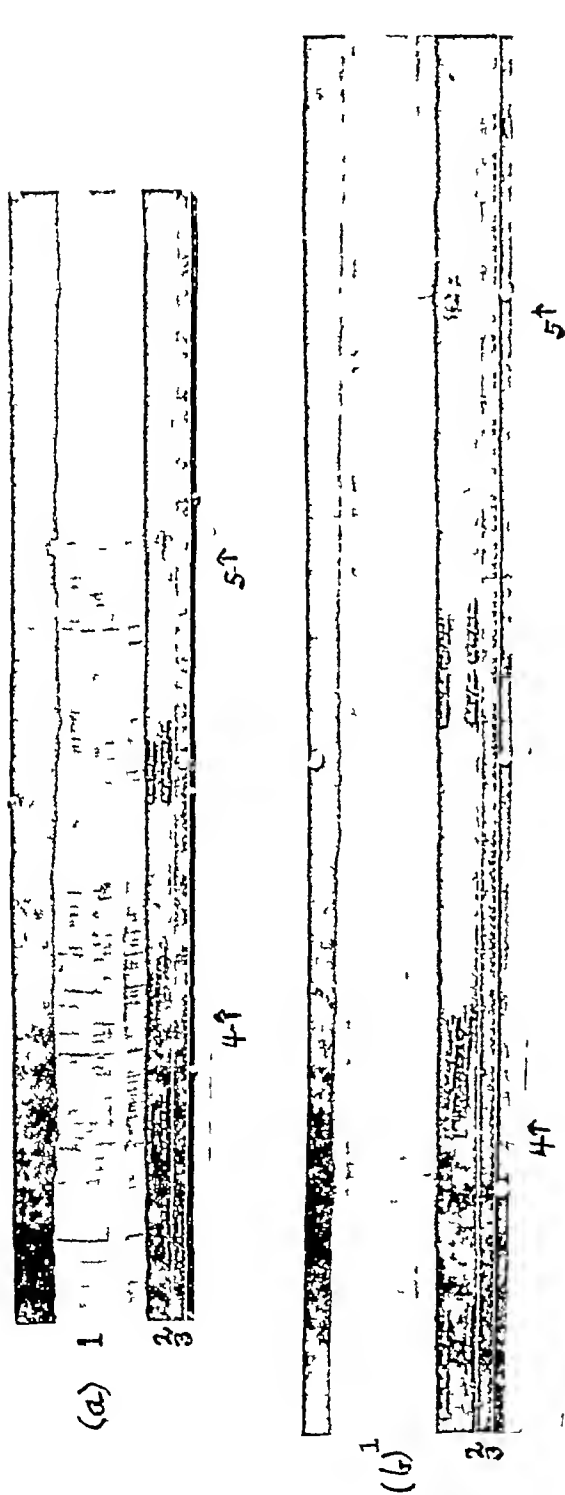


FIG 1 CAFFEIN SODIO-BENZOATE

a, dilution 1-50,000 Increase in coronary flow 5 per cent *b*, dilution 1-25,000 Increase in coronary flow 5 per cent
In all of the figures 1, represents cardiac contractions, 2, coronary flow in which each registration equals 2 cc, 3, time—3-second intervals, 4, administration of drug, 5, the drug discontinued

effect on the cardiac rate. In some instances the amplitude of cardiac contraction was diminished by the drug, associated with some increase in tone (fig 2, *b*).

Theophyllin in concentration of 1-25,000 and 1-50,000 increased the rate of coronary flow from 20 to 45 per cent (fig 3). In some of the experiments there was an acceleration in cardiac rate of approximately 20 beats to the minute. In one instance in which a 44 per cent increase in the rate of the flow of the perfusate was observed, there was

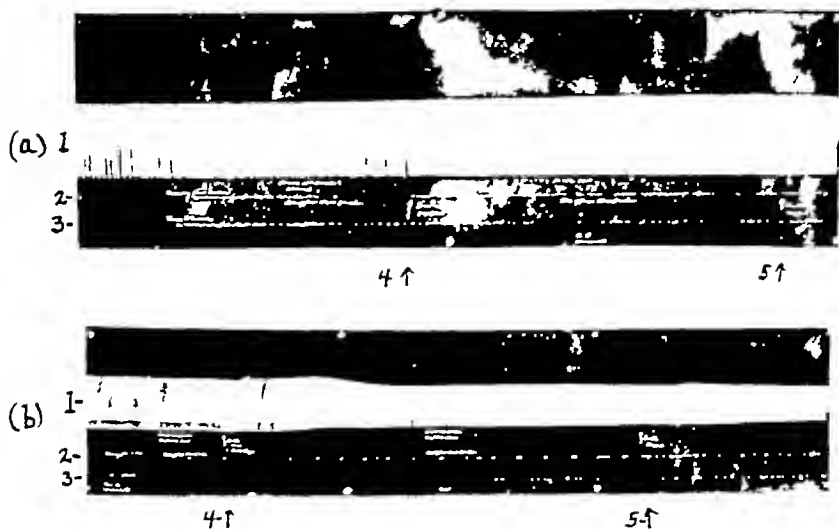


FIG 2 THEOBROMIN SODIO SALICYLATE

a, dilution 1-25,000. Increase in coronary flow 4 per cent. *b*, dilution 1-50,000. Increase in flow 15 per cent.

no acceleration of cardiac rate and no increase whatever in amplitude of contraction. In a few experiments an increase in amplitude of the contraction was observed shortly following the introduction of the drug (fig 3, *c*). This feature was negligible in those instances in which the heart was driven by rhythmically induced break shocks (fig 3 *a*). In these experiments in which a constant heart rate was maintained the influence of the drug on the rate of coronary flow was approximately the same as in those in which a normal cardiac mechanism prevailed (fig 3, *a* and *c*).

Euphyllin in dilutions of 1-25,000 and 1-50,000 increased the rate of coronary flow from 40 to 90 per cent. The drug, in addition, produced a distinct increase in amplitude of the cardiac contractions for a short time following the administration and accelerated the rate from 20 to 30 beats per minute (fig 4, *b* and *c*). The increase in the rate of

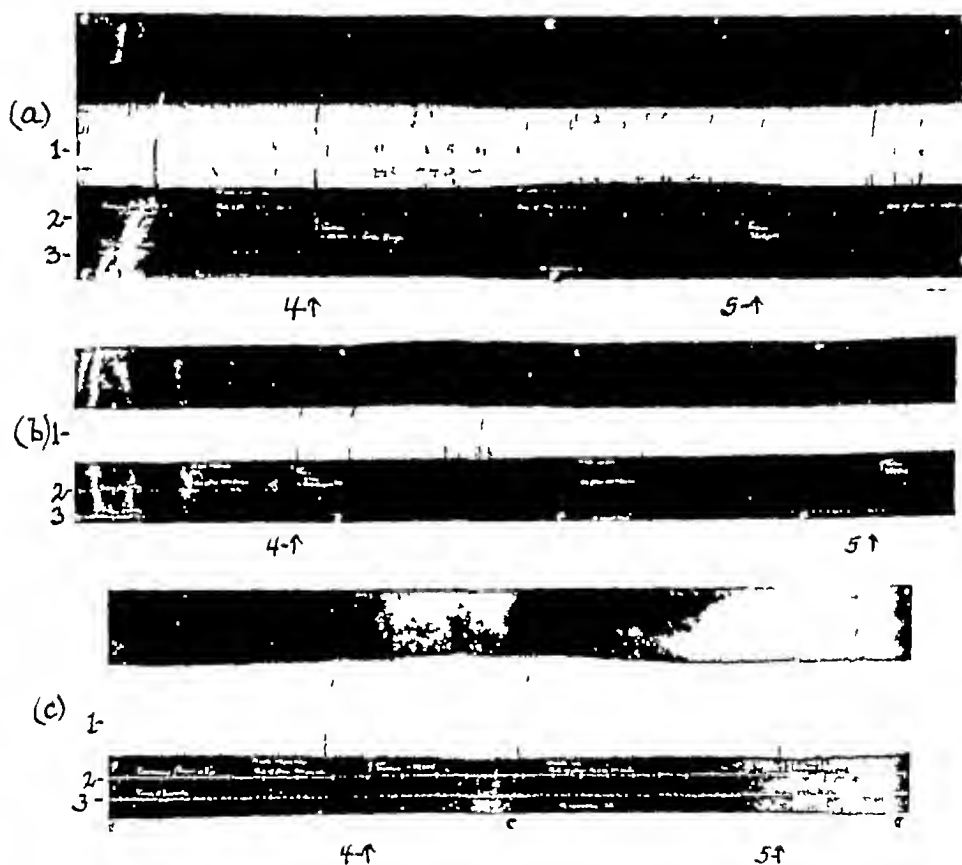


FIG 3 THEOPHYLLIN

- a*, dilution 1-50,000 Increase in coronary flow 27 per cent (Driven heart)
b, dilution 1-25,000 Increase in coronary flow 42 per cent
c, dilution 1-50,000 Increase in coronary flow 28 per cent

the flow of the perfusate was, however, independent of the acceleration in cardiac rate, for practically the same degree of dilating action was observed in those experiments in which the cardiac rate was controlled by electrical stimulation (fig 4, *a*)

In this series of experiments an attempt was made to control, as

far as possible those factors which might influence the rate of coronary flow. It is felt that the changes observed in the flow of the perfusate may in a large measure be attributed to the action of the drug on the coronary vessels.

The action of theophyllin and euphyllin was very striking, whereas that of caffeine sodio-benzoate and theobromin sodio-salicylate was

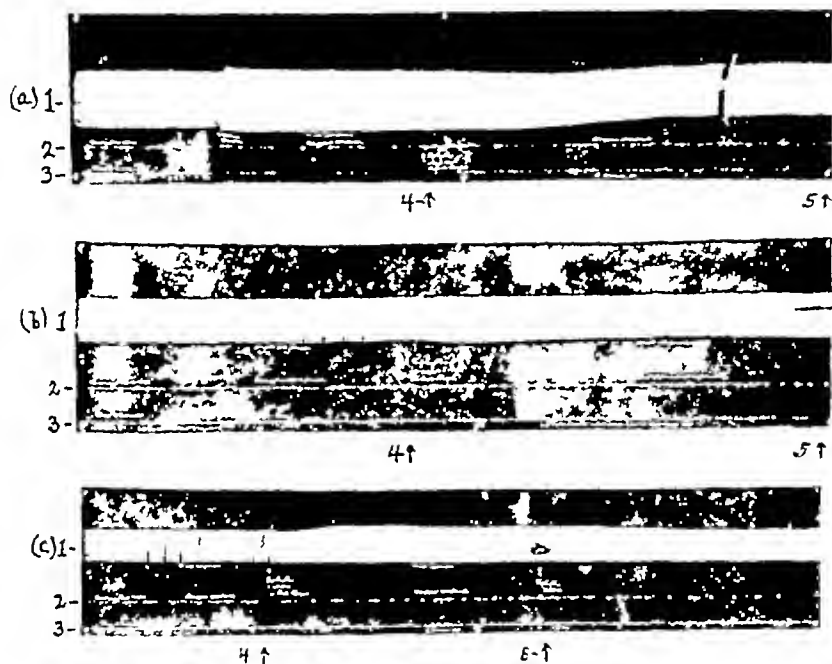


FIG. 4. EUPHYLLIN

a, dilution 1-25,000 Increase coronary flow 60 per cent (Driven heart.)
 b, dilution 1-25,000 Increase in coronary flow 40 per cent c, dilution 1-25,000
 Increase in coronary flow 46 per cent

negligible. The effect of euphyllin was definitely greater than that of theophyllin. These two drugs, in addition to their effect on the flow of the perfusate, exerted a beneficial action on the heart. This feature was not observed following the administration of caffeine sodio-benzoate or theobromin sodio-salicylate. It is to be recalled that the latter drugs, in concentrations of 1-25,000 and even more marked in concentration of 1-12,500, definitely depressed the action of the heart.

The results obtained in this series of experiments from the standpoint of the action of theophyllin and euphyllin on the rate of coronary flow compare very favorably with those reported by Guggenheimer and Sassa. It is to be recalled that they employed similar concentrations of the drug and observed 40 per cent increase in the rate of perfusion with theophyllin and 80 per cent with euphyllin. These authors attributed the augmentation of the rate of the perfusion following the

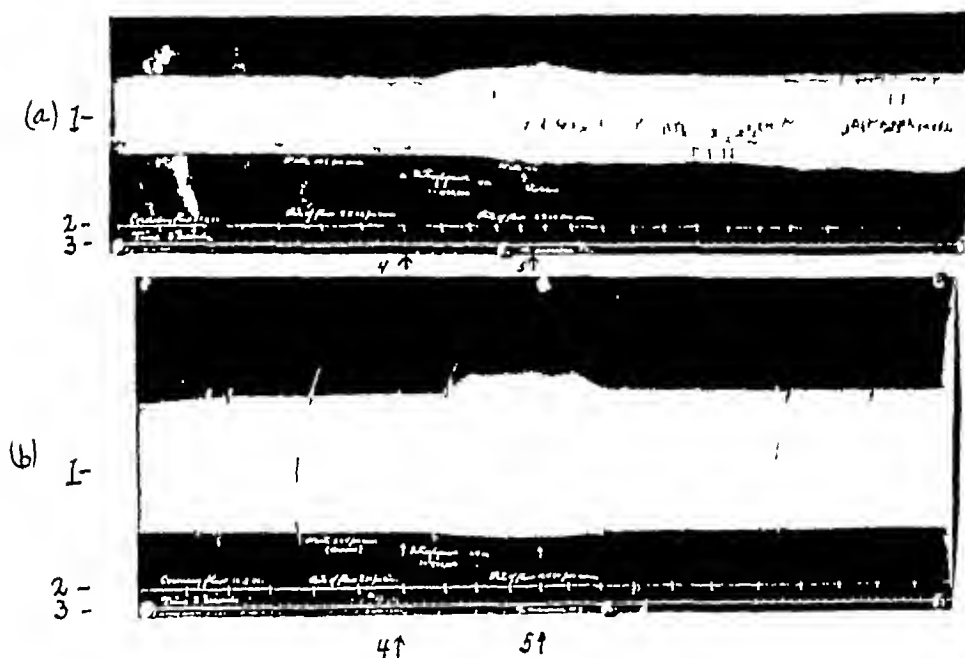


FIG 5 NITROGLYCERIN

Dilution 1-400,000 injected at rate estimated to give concentration of 1-4,000,000 in coronary artery. This calculated to approximate therapeutic dose in man of $\frac{1}{100}$ grain.

a, increase coronary flow 56 per cent. b, increase coronary flow 43 per cent.

administration of theophyllin to the acceleration of the heart rate and the increase in the amplitude of the cardiac contraction. In the present experiments the acceleration of the heart rate by theophyllin was not a prominent feature. It will be recalled that in one instance in which a 44 per cent increase in the rate of coronary flow was observed, the heart rate and amplitude of contraction remained constant. Furthermore, in those experiments in which the heart rate was controlled, practically the same degree of augmentation of the rate of

perfusion was obtained after the introduction of theophyllin. Euphyllin had a somewhat greater tendency to accelerate the cardiac rate. The increase in the rate of perfusion was, however, for the most part independent of this feature as shown in those experiments in which a constant heart rate was maintained by electrical stimulation.

In a few experiments, the spirit of nitroglycerin in dilutions of 1-400,000 and 1-800,000⁴ was employed in order that the action of this drug on the coronary arteries might be compared with that of euphyllin. Nitroglycerin in the above dilutions augmented the rate of perfusion from 40 to 80 per cent, which, as will be recalled, corresponds favorably with that produced by euphyllin. The two drugs produced about the same acceleration in cardiac rate. Nitroglycerin, however, caused a more striking increase in amplitude of cardiac contraction which was maintained for some time after the action of the drug on the rate of perfusion had subsided.

The action of theobromin and theophyllin on the coronary vessels observed in the present investigation was similar to that reported by Heathcote in those instances in which corresponding concentrations of the drugs were employed. A much greater increase in the coronary output was, however, noted by Heathcote when higher concentrations of these drugs were used. In some instances the concentrations of the drugs introduced were as great as 1-2000. In these experiments the great increase in rate of perfusion was probably due to toxic action of the drug on the heart. Furthermore, it is possible that the change in hydrogen-ion concentration produced by high concentrations of the drug might have been a definite factor in producing the marked increase in coronary flow.⁵

CONCLUSIONS

Caffein sodio-benzoate in concentrations of 1-25,000 has little or no effect on the cardiac rate, amplitude of contraction and coronary flow in the perfused heart of the rabbit.

⁴ This concentration was calculated to be comparable to a therapeutic dose of $\frac{1}{100}$ grain in man.

⁵ In an experiment in which the influence of change in the hydrogen-ion concentration on rate of coronary flow was investigated, a decrease in pH from 7.68 to 7.38 increased the coronary flow 7 per cent. When the pH was reduced from 7.68 to 7 the coronary flow was augmented 80 per cent.

Theobromin sodio-salicylate in concentrations of 1-25,000 produces no change in rate of coronary flow. In higher concentrations, as 1-12,500, the rate of perfusion was decreased. The drug apparently had no effect on cardiac rate. In some instances the amplitude of contraction was diminished.

Theophyllin in concentrations of 1-25,000 and 1-50,000 augments the coronary flow from 20 to 45 per cent. The acceleration of the cardiac rate by the drug is not a prominent feature. The greatest increase in coronary flow was produced by the drug when there was no change in cardiac rate.

Euphyllin in above concentrations increased the rate of perfusion from 40 to 90 per cent. Euphyllin has a somewhat greater tendency to accelerate cardiac rate than theophyllin. With both drugs, however, the increase in coronary output was independent of the accelerating action on the heart, as was indicated by experiments in which a uniform cardiac rate was maintained by rhythmical stimulation.

The action of euphyllin on the coronary flow compares favorably with that of nitroglycerin when comparable concentrations of the drugs are introduced into the perfusate.

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TOTAL ACID-BASE EQUILIBRIUM OF PLASMA IN HEALTH AND DISEASE

VI STUDIES OF DIABETES

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(Received for publication August 6, 1925)

INTRODUCTION

In the first article of this series (1) a method for the determination of the total acid-base equilibrium of serum was presented together with certain studies of the electrolyte equilibria of healthy persons under various circumstances and of hospital patients who presented miscellaneous conditions that disturbed the electrolyte equilibrium of the blood. The most important point established by these studies was the fact that the organism responds to the production of abnormal amounts of acids of various kinds or the reduction of bicarbonate by a transfer of water, chloride or base from the tissues to the blood or vice versa. Reduction of the plasma chloride by this means liberates base for combination with other acids and, therefore, tends to reduce acidosis, while an increase of chloride has the opposite effect. A transfer of chloride in one direction is equivalent to a movement of base in the opposite direction. The whole mechanism confers on the blood a greater ability to resist the disturbing effects of conditions that alter electrolyte equilibrium by rendering the acid and base stores of the tissues available where and when they are most needed. The reactions involved must be quite comparable to those which have long been known to occur between the cells and the plasma in response to similar chemical disturbances. They may occur with such rapidity as to produce recognizable differences in the composition of arterial and venous blood and their ability to absorb, transport and dissociate carbon dioxide and oxygen.

ACID-BASE EQUILIBRIUM VI DIABETES

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Case number	Hospital number	Date	1	2	3	4	5	6
			Oxygen capacity	Cell volume	Ratio of column 1 to column 2	Proteins	CO ₂	
			vols per cent	vols per cent		per cent	vols per cent	
1	5105	November 23, 1922	21 3	45 6	46 7	6 91	54 5	30
2	15533	January 30, 1923	21 1	43 9	48 0	7 66	51 5	30
3	Alf	June 4, 1924						
4	39423	January 4, 1925				6 76	66 6	33
5	10888	November 22, 1922	17 8	38 3	46 6	6 47	74 2	30
6	12562	November 7, 1922	20 0	43 0	46 5	7 41	48 9	45
7	18582							
8	16727	February 16, 1923	20 5	44 6	46 0	7 36	57 5	34
9	29843	April 5, 1924						
10	22718	June 2, 1924						
11	33528	June 11, 1924						
12	20387	January 17, 1924	18 1	38 2	47 4	5 39	57 2	33
13	33331	May 25, 1924						
		May 31, 1924						
14	33222	May 11, 1924						
		May 31, 1924						
15	33229	May 13, 1924						
16	10801	November 17, 1922	19 5	41 7	46 9	6 60	45 1	30
17	29771	April 1, 1924						
		May 23, 1924						
18	26315	November 9, 1923					61 4	34
19	15733	February 20, 1923	17 8	38 5	46 1	5 98	63 8	34
20	34490	September 5, 1924						
21	33333	May 23, 1924						
		June 2, 1924						
22	34618	September 19, 1924	17 7	34 2	51 8	6 40	58 9	35
23	34156	July 31, 1924						
24	19838	June 16, 1923	20 1	41 5	48 4		53 3	30
25	18867	June 11, 1923	18 3	39 7	46 1		41 2	34
26	15267	January 3, 1923	19 2	37 4	51 4	6 28	52 8	31
27	15924	May 9, 1923	14 3	32 3	44 3	6 25	43 5	35
28	15175	November 29, 1924	20 9	41 9	49 9	7 25	53 4	35
29	10572	October 24, 1923	17 5	41 4	42 2	7 68	45 1	36
30	18467	November 7, 1923	15 3			6 72	59 2	37
		November 7, 1923	16 6			6 26	55 4	38
31	29754	March 28, 1924	20 1	44 0	46 0	7 93	43 9	39
		March 30, 1924	17 7	40 7	43 3	6 66	85 0	24
32	36016	February 21, 1925		40 6		6 07s	78 0	38
33	10629	October 26, 1922	18 7	41 5	45 1	7 48	47 8	37
		October 30, 1922					46 1	38

PETERS BULGER EISENMAN AND LEE

7	8	9	10	11	12	Blood sugar	Nature and treatment of blood	Urine sugar	Urine acetone
mg P	Total acid	Total base	Organic acid	pH	Blood non protein nitrogen				
per cent	mlf	mlf	mlf		mg per 100 cc	mg per 100 cc			
	146 8			7 36		231	\ cap	0	0
	143 9			7 33	29	119	\ cap	+	+
3 7		161 7			37	172	\ cont.	0	0
	142 9					138	\ cont.	0	0
	147 2			7 50	33	311	\ cap	0	0
	157 9			7 31	30	139	\ cap	0	0
		178 8			27	271	\ cont.	0	0
	147 3			7 38		147	\ cap	0	0
		165 0			30	163	\ cont.	0	0
3 6		162 2				126	\ cont.	0	0
4 8		165 5				185	\ cont.	0	0
	136 6			7 38	33	263	\ cap	+	0
2 6		153 3				251	\ cont.	0	0
2 9		166 0				111	\ cont.	0	0
		160 1			32	230	\ cont.	2+	0
4 1		160 3				169	\ cont.	0	0
		162 1			29	231	\ cont.	4+	0
	140 5			7 27	31	411	\ cap	3+	0
		161 1			28	266	\ cont.	4+	0
4 4		173 0				160	\ cont.	0	0
	135 2			7 47		316	\ cont.	4+	0
	142 4			7 43	30	319	\ cap	4+	±
2 7		137 4				217	\ cont.	2+	0
		166 6				188	\ cont.	2+	0
3 9		151 9				185	\ cont.	0	0
	138 6					309	\ cont.	4+	4+
3 3		147 6			28	155	\ cont.	4+	4+
	131 3			7 39		220	\ cont.	4+	+
	126 0			7 39		326	\ cont.	2+	+
	126 6			7 34		811	\ cap	4+	3+
	118 2				65	112	\ cont.	0	0
	134 4			7 35	37	244	\ cap	0	0
	131 6			7 27		271	\ cap	4+	4+
3 1	136 9			7 51		322	\ cont.	4+	0
3 6	135 1			7 42		133	\ cont.	0	0
2 6	130 0					234	\ cont.	4+	4+
2 4	135 4			7 44		113	\ cont.	0	+
3 0	131 9	141 3	9 4		33	226	\ cont.	2+	+
	139 4			7 30		291	\ cap	4+	3+
	142 8			7 28		366	\ cap	4+	3+

ACID-BASE EQUILIBRIUM VI DIABETES

THE

Case number	Hospital number	Date	1	2	3	4	5	6
			Oxygen capacity	Cell volume	Ratio of column 1 to column 2	Proteins	CO ₂	O
			vols per cent	vols per cent		per cent	vols per cent	per cent
34	29127	February 22, 1923	18 4	42 6	43 2	6 72	57 4	28
35	35283	November 29, 1924	25 2	59 8	42 1		59 9	37
36	34704	September 25, 1924					19 5	34
37	15120	December 13, 1922	20 1	46 2	43 4	7 14	50 6	33
		December 14, 1922	22 3	46 8	47 6	7 43	52 1	30
38	33964	July 12, 1924	16 1	39 6	40 7	6 64	54 2	316
39	18067	March 21, 1923	23 3	44 7	52 0	5 45	16 5	342
40	34878	October 16, 1924	17 6	36 7	47 9	5 69s	40 3	341
41	34513	October 25, 1925	19 7			6 03	51 3	330
		October 27, 1925				4 57	57 6	322
42	35857	February 8, 1925	20 4	49 6	41 1	6 24s	66 6	349
43	34611	September 17, 1924				6 13s	41 4	
		September 22, 1924	19 6	42 7	45 9	5 45s	63 0	336
44	16670	February 12, 1923	13 8	27 8	49 8	5 36	30 9	310
		February 22, 1923	15 2	30 9	49 0	5 03	62 1	372
		August 25, 1923					26 6	331
		November 3, 1924						
45	10572	November 14, 1922	17 2	36 2	47 6	5 75	63 3	360
46	34295	August 23, 1924	18 0	41 5	43 4	6 22	70 3	358
		August 30, 1924	16 7			6 64	68 0	355
47	36140	March 4, 1925	19 9			6 39s	22 0	
		March 6, 1925	18 3	43 4		5 22s	50 6	351
		March 14, 1925	16 2			4 95s	64 7	315
		March 23, 1925	14 8			4 98s	61 8	389
	36140	March 31, 1925	17 4			6 17s	67 6	375
48	15096	December 11, 1922	22 4	47 2	47 4	7 29	51 1	275
		December 13, 1922	21 3	45 1	49 3	5 77	76 5	211
		January 11, 1923	18 4	40 1	45 9	6 30	54 1	388
49	22513	August 16, 1923, a m					36 8	340
		August 16, 1923, p m					36 6	361
		August 17, 1924					41 2	354
		August 18, 1923					39 5	353
		August 19, 1923					41 3	321
		August 20, 1923					45 6	352
		August 21, 1923					52 3	351
50	29061	January 28, 1924	22 5	45 8	49 3	8 37	16 8	355
		January 29, 1924	21 0	42 4	49 6	8 07	41 6	
		January 30, 1924	19 0	40 6	46 8	6 20	49 3	39
		February 6, 1924	18 4	38 2	48 2	5 85	60 9	3

8	9	10	11	12	Blood sugar	Nature and treatment of blood	Urine sugar	Urine acetone
Total acid	Total base	Organic acid	pH	Blood non protein nitrogen				
mM	mM	mM		mg per 100 cc	mg per 100 cc			
121 3			7 61	33	167	\ cont	4+	0
145 4	178 2	32 8		53	88	\ cont.	4+	2+
112 8	132 3	13 5			469	\ cont	4+	4+
129 9			7 32	39	449	\ cap	4+	2+
123 1			7 34		274	\ cap	0	0
142 6	168 2	25 6		54	255	\ cont.	4+	+
111 8			6 78	38	1,000	\ cap	2 5%	4+
124 4	156 3	31 9		25	206	\ cont.	2+	4+
126 1	161 9	35 8		43	197	\ cont	4+	2+
124 8					176	\ cont	+	0
139 4	181 8	42 4		55		\ cont	2+	+
					315	\ cont	4+	3+
133 6	149 1	15 5	7 48		341	\ cont.	0	0
127 4			7 09		441	\ cap	4+	4+
142 1			7 42		313	\ cap	+	+
118 0			7 02		435	\ cap	4+	4+
	162 7					\ cont.	0	0
142 2			7 43		290	\ cap	0	+
143 9	144 6	0 7			200	\ cont.	2+	+
143 3	144 7	1 44	7 39		152	\ cont.	0	0
146 6	146 6			37	341	\ cont.	4+	3+
130 3	153 4	23 1		24	179	\ cont.	+	+
144 1	163 0	18 9			127	\ cont.	0	0
144 4	156 3	11 9			155	\ cont.	+	0
147 0	155 4	8 4		23	206	\ cont.	0	0
114 5			7 33	67	742	\ cap	4+	4+
123 5			7 51	39	216	\ cap	0	0
145 0			7 35	22	297	\ cap	0	+
127 2			7 18	44	497	\ cap	4+	4+
129 9			7 17		400	\ cap	4+	4+
130 4			7 23		278	\ cap	4+	4+
129 2			7 21		294	\ cap	4+	4+
121 3			7 23		251	\ cap	4+	4+
131 9			7 28	37	233	\ cap	4+	4+
131 1			7 34		200	\ cap	3+	3+
118 5			7 28	37	308	\ cont.	4+	4+
			7 37		142	\ cont.	4+	+
144 7			7 52		229	\ cont.	+	0
138 8			7 54		241	\ cont.	+	0

Case number	Hospital number	Date	1	2	3	4	5
			Oxygen capacity	Cell volume	Ratio of column 1 to column 2	Proteins	CO ₂
			rols per cent	rols per cent		per cent	rols per cent
51	22350	November 19, 1923	20 1			8 02	13 5
		November 20, 1923	20 1			7 45	33 7
		November 21, 1923	17 0			5 94	47 8
		November 27, 1923	16 9			6 24	73 8
		December 17, 1923	16 7			6 73	67 4
		September 6, 1924					19 0
		September 7, 1924				5 06s	26 7
		September 8, 1924				3 22s	47 0
		September 10, 1924				4 75s	28 0
		September 16, 1924				6 70s	59 8
		September 24, 1924	16 5	37 6	43 9	6 14s	57 7
		October 7, 1924	16 5	40 9	40 4	6 98s	65 1
52	29176	February 9, 1924	18 9	40 6	46 6	6 95	20 8
		February 10, 1924	16 7	36 8	45 1	6 37	38 3
		March 5, 1924	14 8	34 2	43 3	6 23	66 2
		November 10, 1924, 2 p m	20 8	58 2	35 7	7 74s	14 5
		November 10, 1924, 10 p m	20 5	51 2	40 0	7 60s	19 5
		November 11, 1924	20 3	48 4	41 8	6 90s	43 1
		November 13, 1924	16 9	43 8	38 6	6 55s	55 6
53	26416	March 26, 1924	20 4	44 3	46 1	6 37	56 9
		March 31, 1924	19 2	42 5	45 2	6 20	64 0
		March 22, 1925	20 0	48 5	41 2	7 43s	7 6
		March 24, 1925	20 1	46 2	43 5	7 40s	43 0
		March 27, 1924	15 4	37 7	40 9	6 03s	51 7

chloride after the administration of glucose that support such a theory. One of the authors³ several years ago attempted to establish a similar relation between blood sugar and plasma chlorides unsuccessfully. In certain experiments reciprocal variations of the two elements were observed, but this effect could not be produced consistently. The

³ Peters, J. P. Unpublished experiments.

8	9	10	11	12	Blood sugar	Nature and treatment of blood	Urine sugar	Urine acetone
Total acid	Total base	Organic acid	pH	Blood non protein nitrogen				
mM	mM	mM		mg. per 100 cc.	mg. per 100 cc.			
			7.07	37	616	V cont.	0.8%	4+
127.2			7.31		344	V cont.	2.0%	3+
131.5			7.30		349	V cont.	0.3%	0
134.8			7.45		249	V cont.	+	0
137.0			7.52		242	V cont.	0	0
143.1								
	143.1			31	462	V cont.	2.1%	3+
	152.5	28.9		37	370	V cont.	2.5%	4+
123.6	150.7	20.8		37	233	V cont.	2.3%	4+
129.9	152.6	45.3		35	257	V cont.	2.5%	4+
106.8	140.5	-3.5		29	270	V cont.	1.1%	+
144.0	145.8	7.0	7.49		462	V cont.	0	0
138.8	154.7	9.2		22	130	V cont.	0	0
145.5								
			7.29		300	V cont.	4+	4+
117.2					121	V cont.	0	2+
127.0			7.34		150	V cont.	0	0
147.3								
	150.5	37.8	6.95		317	V cont.	3.5%	4+
112.7	143.7	32.8			429	V cont.	3.5%	4+
110.9	143.6	10.3	7.29		278	V cont.	5.5%	
133.3	140.8	5.7			246	V cont.	2.8%	0
135.1								
			7.19		263	V cont.	4+	4+
133.6			7.29		199	V cont.	+	2+
134.0								
	154.2	34.7		46	450	V cont.	4+	4+
119.5	146.5	11.8		37	174	V cont.	2+	+
134.7	140.6	3.1		29	283	V cont.	3+	0
137.5								

administration of large amounts of glucose causes such a variety of changes in the metabolism and the composition of blood that a study of something more than the chlorides is necessary before one can discuss osmotic equilibrium. Although total base is not in any sense a measure of the osmotic pressure of the blood, it does offer an estimate of the fraction of the total osmotic pressure contributed by the electro-

lytes If the electrolytes compensate for the accumulation of non-electrolytic osmotically active substances such as glucose, one would expect to find some evidence of such a mechanism in diabetic patients with high blood sugar On the contrary, in the cases of our series with hyperglycemia, but without ketosis, the level of base is high rather than low In some cases, to be sure, such as 13 and 17 base and glucose change in opposite directions On the other hand, in 14 base remains unchanged as the blood sugar falls and in 21 base falls to a surprising extent while the level of glycemia remains entirely unaltered

Examination of the different acids in the 10 cases in which they were studied shows, with two exceptions, a rather high total acid level, which is due usually to an elevation of chloride (the high CO_2 of 5 followed the administration of salicylates and sodium bicarbonate)

Although the general trend of both acid and base figures is towards high rather than low values, there are several exceptions to the rule so striking that generalizations concerning the character of the electrolyte picture of diabetes seem hardly warranted If one examines the protocols of these patients which are presented in brief below, one can hardly be surprised that such variations are encountered The average diabetic patient who enters a general hospital does so not because he has diabetes, but because of some concomitant condition which has usually aggravated the diabetes The effect of these conditions must be taken into account in any analysis of blood chemistry Further than this the manifestations of uncomplicated diabetes are variable Diuresis or polyuria and other symptoms do not always bear a constant relation to the disorder of carbohydrate metabolism This is well illustrated by the findings in 12 Proteins and chloride were both low in this case although there was nothing peculiar in the clinical picture Careful study of the clinical notes revealed the fact that on the day the blood was taken she developed a striking diuresis that continued for 3 days

Cases 22 to 26 represent 5 observations on patients with uncomplicated diabetes of a severe type with marked ketosis, but without any dyspnea and without any considerable reduction of plasma CO_2 To these may be added the first two observations of 54, the last case in the table The dissociation of acidosis and ketosis has long been

recognized Ketonuria without reduction of the alkaline reserve is not uncommon in patients with moderately severe, uncomplicated diabetes Y Henderson (15) indeed uses this as an argument against the current theory of diabetic acidosis which presumes that the base which neutralizes ketone acids in the blood is derived from bicarbonate He says "if lactic acid or any other strong acid is added to blood, most of the alkali which neutralizes it is drawn from the hemoglobin alkali reserve, and it causes, therefore, only a very slight decrease in the plasma bicarbonates" It has already been shown (1) that in the case of the lactic acid of exercise, to which Henderson is especially referring in this sentence, most of the alkali which neutralizes the foreign acid is derived not from the hemoglobin, but from the tissues In this case, however, the body is dealing with an acid that can be and usually is oxidized The provision of alkali from the tissues does not, therefore, ultimately lead to any appreciable depletion of the base stores of the organism The acidosis of diabetes, on the other hand, is due to the accumulation of acids which the body can not destroy and which can only be disposed of through the kidneys in combination with a certain amount of base From the diminution of the chlorides exhibited by these patients it would appear that in conditions of more or less chronic ketosis these acids were able to spare bicarbonate, robbing chloride of its base instead This not only preserves the bicarbonate, but also prevents a change of blood pH

On the other hand it may be that the chloride recession is only a means of maintaining a fixed level of base and a constant electrolytic concentration From these experiments alone it is impossible to decide between these alternative possibilities because there are no simultaneous determinations of acid and base In the case in which base was determined it proved to be quite low In this instance, certainly, the electrolytic fraction of the osmotic pressure was low Whatever may be the mechanism which determines the depression of chloride, the base which neutralized the bicarbonate was certainly not derived, as Henderson suggests, from the hemoglobin The base combined with hemoglobin is, as Van Slyke, Wu and McLean (3) have shown, determined by the pH and the concentration of hemoglobin in the cells The pH of the cells bears a definite relation to that of the plasma The latter, in these cases is normal as is the height

of the carbon dioxide absorption curve. The amount of base combined with hemoglobin was also, then, presumably normal because it is inconceivable on theoretical grounds that the carbon dioxide and pH of the cells could have been reduced without affecting the plasma. That there may be no possible doubt about the matter, however, it may be mentioned that the whole bloods of 24, 25 and 26 were also analyzed for CO_2 . The relation between the CO_2 of cells and plasma was in each case quite normal.

The majority of the cases in the table from 27 on demonstrate quite conclusively the point made above, that in estimating the cause of changes in the metabolic or chemical picture of diabetic patients due attention must be given to the associated pathologic conditions. Clinical knowledge of diabetes would be measurably advanced if case reports and studies contained more details regarding the clinical condition of patients, even at the expense of metabolism data. This series of cases gives a fairly representative idea of the type of conditions which the severe diabetics in a general hospital present. There has been much discussion of Bock, Field and Adair's (16) discovery of organic acids other than ketone acids in diabetic patients. There is no reason to consider this as extraordinary unless it can be proved that these acids were produced as the result of the metabolic disorder of diabetes itself. This they did not prove. Case 4 of their series presented symptoms not usually encountered in uncomplicated diabetes and a well marked elevation of temperature. In our experience diabetic acidosis and coma may be taken as presumptive evidences of some infection or other pathologic condition and are seldom, if ever, as is generally taught, entirely the result of mismanagement or dietary indiscretions. The latter bring their own uncomfortable rewards of a different kind and may, to be sure, greatly intensify the harmful effects of any accident or infection. Often enough the infection is so slight that it is overlooked or neglected. Case 44 shows what even a slight cold may do to a diabetic patient who observed dietary regulations at all times with meticulous care. On two occasions a mild coryza precipitated severe ketosis and acidosis and brought him to the hospital on the verge of coma.

Cases 27 to 32 demonstrate that low chlorides are often observed in diabetic patients without ketosis and in the presence of a normal

alkaline reserve This does not mean that the reductions noted in the previous group may not have been at least in part, due to ketosis Some light is thrown on this question by the contrast between 30 and 31 On both two observations were made one at the time of admission when there was an extreme hyperglycemia and glycosuria, the second when both hyperglycemia and glycosuria had been reduced by treatment Case 31 also had severe ketosis at the time of his first examination, while 30 showed no ketonuria The total acid concentration of the latter remained unaltered after treatment, while that of 31 increased, presumably to satisfy the base freed by the combustion of organic acid

The mechanism of the acid increase is interesting As chloride had been largely responsible for the original low acid level, one would naturally have expected chloride to play the major part in the restoration of equilibrium On the contrary chloride instead of going up fell further, while bicarbonate rose to an excessively high point This tendency for bicarbonate to overshoot the mark in the recovery from acidosis was first pointed out by one of the authors (17) in 1917 and has since been confirmed by Stillman Van Slyke, Cullen and Fitz (18), Cullen and Jonas (27) and Bock Field and Adair (16) The latter have, furthermore, found that this rise of bicarbonate results in shifting the pH somewhat to the alkaline side of normal It had already been shown by Peters (17) and by Stillman et al (18) that the alveolar CO_2 under these conditions remained relatively low, which in itself was an indication that such a shift of pH must occur

A possible explanation of this phenomenon is offered by these experiments The low base of cases 23 and 32 and the general low level of total acid and chlorides even when there is no ketosis and therefore, in all probability, no great excess of organic acid, intimates that the blood, at least, is deficient in both chloride and base It is, perhaps, advancing too far to suggest that this deficiency in the blood may reflect a similar depletion of the tissues If such a depletion does exist, however, it is not impossible that it has its origin in an excessive excretion of chloride From the standpoint of the blood alone, in any case, chloride does not appear to be available to replace the base released by the combustion of the ketone acids Carbon dioxide has, therefore to bear the whole load CO_2 is, however, rendered

available only as it is produced in the metabolism and excretion usually follows production *pari passu*. The rapid seizure of carbon dioxide by the newly liberated base reduces the tension of free carbon dioxide and consequently the pulmonary carbon dioxide excretion, and causes the pH of the blood to rise.

If there is a depletion of the chloride and base stores of the body this depletion is not a direct result of ketosis because it is encountered when ketonuria is absent. Furthermore, there is no reason to suppose that the excretion of ketone acids, which in themselves abstract base from the tissues, would provoke a chloride diuresis which could only withdraw further base at a time when it is most urgently required. Diabetic polyuria may lead to the excretion of excessive chloride. Unfortunately no opportunity to study the inorganic metabolism of these patients during the development of the acute condition is afforded, especially since in insulin such an efficient weapon has been provided against all the misfortunes of diabetes. Serious diuresis seldom survives efficient insulin therapy long. The comparatively low chloride, protein and total acid of 12, mentioned above, which coincided with the institution of a secondary polyuria would be readily explained if diabetic polyuria is associated with chloride diuresis.

On the whole, it is evident, that the majority of patients with uncontrolled diabetes of a type inherently severe or rendered serious by some concurrent condition, whether they develop ketosis or not, generally exhibit a reduction of plasma chlorides and probably of total base. Case 33 is an exception, but it is hard to estimate in this case the relative effects of diabetes, fractured skull and therapeutic measures.

Cases 34 to 38 had pneumonia. Cases 34, 36 and 37, with typical lobar pneumonia, presented extremely low chlorides and total acid, even in the absence of ketonuria. If diabetes and pneumonia both tend to reduce chloride, such a result might reasonably be expected. The pneumonia of 35 certainly began as a pulmonary infarction. This, the presence of severe cardiac decompensation and the previous subcutaneous administration of saline may together account for the high base, acid and chloride.

This experiment is the first in which ketonuria occurs and in which both total base and acid were determined. It affords, then, the first opportunity for the estimation of the value of the procedure as a

means of detecting and evaluating the accumulation of ketone acids in the blood. The value of organic acid is, as it should be, far above normal. By the same criteria, however, the method fails completely in the next case, 36, in whose serum only 13.5 millimols of organic acid were found although she was in diabetic coma and had marked ketonuria. This stands as the only observation in which ketonuria was discovered without a concomitant increase of the organic acid of the serum. The accuracy of the procedure must not be lightly condemned on this account. In the first place the proteins were not determined and the value ascribed to them was probably excessive because it is fair to assume in the light of experience that pH and protein concentration were both reduced.

Case 38 had high base, normal acids and plasma organic acid high in proportion to the degree of ketosis indicated by urine examination. She had, however, advanced cardiac decompensation, edema and almost complete anuria. Severe ketonemia with minimal ketonuria as a terminal event in diabetes has been encountered not infrequently. On the other hand it is quite as possible that the organic acidemia was caused by other acids, the product not of diabetes but of the associated diseases.

Case 39 had been vomiting for about 36 hours before she was admitted to the hospital. Vomiting, with acute abdominal pain as the precipitating cause of diabetic toxemia appears several times in this series of patients. In this case and in that of 53 the picture was so strongly suggestive of an acute abdominal condition that the advisability of immediate exploratory operation was seriously considered. In most instances rest, proscription of fluids by mouth, the administration of large amounts of fluid containing salt and glucose by other routes together with large doses of insulin has resulted in the rapid elimination of symptoms. The improvement is so rapid in these cases that it is hard to believe that the syndrome is referable to organic disease of the pancreas or any other abdominal organ. Case 52 both times that she was admitted presented a similar syndrome. In her case it seemed quite possible that the attacks were initiated by some disorder of the action of the heart.

The electrolyte disturbance produced in the diabetic by vomiting is quite different from that which develops in the normal individual

Plasma chloride does, to be sure, diminish and base may, as will appear subsequently, also fall. Bicarbonate decreases, however, with extraordinary rapidity and serious acidosis may eventuate within a few hours. Either the vomiting or the underlying cause of the vomiting has a profound effect on carbohydrate metabolism, practically deleting the power of the individual to utilize glucose. This is evidenced by the maximum blood sugar values encountered in vomiting cases. The inevitable consequence of this loss of carbohydrate tolerance is the rapid accumulation of ketone acids in the blood. This may be furthered by the failure of the excretory powers of the kidney to which the authors have called attention in another connection (25). The case is comparable to the acidosis which develops in vomiting infants. Shaw, Moriarty and Talbot (20) have shown that this acidosis is due to a starvation ketosis and may be rapidly relieved by the administration of carbohydrate. They believe that ketosis can be produced more easily in infants than in adults because the former possess a smaller reserve supply of carbohydrate. To this must be added another factor. As vomiting reduces the chloride and base of blood the usual defenses of the body against organic acid is diminished. Bicarbonate must, as it were, bear the whole acid burden alone. This leads to earlier alteration of pH in the acid direction.

The effects of intestinal obstruction and peritonitis attended by vomiting were briefly presented in the preceding paper of this series (1). Chloride and base proved to be reduced. Case 40 presented constipation of such severity that it amounted to complete intestinal obstruction. For three days preceding the examination her bowels had not moved. She had not vomited, but had taken little food. The diabetes had been so aggravated that there was considerable ketonuria, but total base had not diminished. Chloride is, indeed, reduced and bicarbonate has also suffered some loss, but only because they have been displaced by organic acid. Case 41, with cholelithiasis presents a very similar picture, she had vomited only once during the course of the abdominal condition.

The next case, 42, does not properly belong among true diabetics. With an intestinal obstruction following resection of the cecum he developed glycosuria, hyperglycemia and ketonuria. The metabolic disturbances disappeared completely shortly after the operation. At

the time of the blood examination he had received insulin and glucose, but glycosuria and ketonuria had not yet cleared up. This explains the high organic acid concentration of the serum. The excessive amount of base may be the result of large hypodermic saline injections which he had received. If this is the case the low chloride must be looked upon not as a true salt deficiency, but as a displacement of chloride, probably into the tissues, to free base to the ketone acids.

Case 43 illustrates the electrolyte picture during the latter stages of the recovery from severe acidosis provoked by an attack of vomiting. Base is still somewhat depressed, although bicarbonate has returned to the normal level, chloride is quite low. The pH is near the upper limit of normal.

The occurrence of edema in diabetic patients has long been recognized, but has never been explained in a satisfactory manner. Because it can be delivered rapidly by the administration of acidifying diuretics (21, 22) and because it seldom if ever, appears in association with acidosis it is generally believed to be in part due to an alkalosis. The six patients, 44 to 49 inclusive all presented subcutaneous edema at some stage in the course of their illnesses. All attempts to connect the edema with a characteristic electrolyte picture fail. It is quite true that edema never occurred during acidosis, but the level of bicarbonate during edema was frequently well below the upper normal limit. That acidosis causes dehydration and recovery from acidosis is attended with the retention of water is well established (23). During the recovery from severe diabetic acidosis body weight increases at a surprising rate and to a remarkable extent even if edema does not develop. There is less certain evidence that the administration of excessive amounts of alkali will produce an abnormal retention of fluid. In a healthy individual it has not proved possible to provoke a true edema by means of alkali. In certain pathologic conditions, however, edema has been produced by the administration of sodium bicarbonate, as if the disease had increased the sensitivity of the organism to the hydrating effect of alkali. Thus Binger, Hastings and Neill (24) have reported the occurrence of edema after bicarbonate therapy in a case of pneumonia.

Under certain circumstances diabetes seems to confer such a sensitivity to alkali. Case 49 developed a moderate edema, in spite of

the presence of severe ketosis, after she had received sufficient bicarbonate to restore the carbon dioxide absorption curve of the blood to the normal level. In another connection (19) it was pointed out that this edema did not affect the blood which was highly inspissated as is indicated by the high hemoglobin, cell volume and plasma protein. It did succeed in displacing chloride, which was probably already low because of the combination of ketosis and vomiting, to an extreme degree. By the administration of insulin ketosis was entirely eliminated in the course of 36 hours and the base released by the ketone acids was taken up by bicarbonate. As the latter had been high at the outset it rose far above the normal limit. The total acid, however, remained low and vomiting persisted. With the impression that the chlorides and base of the body were depleted the patient was given sodium chloride on the third day. This was followed almost immediately by alleviation of symptoms. With a clear recognition of the dangers of *post hoc* reasoning the results of sodium chloride therapy in this and other similar cases has seemed to the authors to be of distinct benefit, a further intimation that the low chlorides and base indicate salt depletion.

Comparison of the other four cases during and subsequent to edema shows that the latter may occur when base is high or low, when chloride is high, low or normal, when protein is at any level. Finally, from 46 it appears that it may come and go without any significant change of the acids or base. The ultimate cause of the predisposition to the hydrating effect of alkali which these diabetic patients exhibit the authors are inclined to ascribe to malnutrition. In their experience edema is encountered only in patients who have undergone tissue wastage as a result of severe or improperly treated diabetes. In all the cases they have encountered edema has disappeared spontaneously when an adequate diet had been maintained for a sufficient period. If this is the case diabetic edema, for which no cause can be found in complicating pathologic conditions may be accepted as evidence that the patient has received an inadequate diet. At the same time it is a presumptive indication that he is not suffering from acidosis.

The next case, 50, illustrates again the manner in which chloride comes to the aid of bicarbonate in compensating chronic ketosis.

The same is true of 51. In this case feeding was difficult because of the mental condition of the patient. Whether it was failure to administer sufficient salt or the development of pneumonia that caused the reduction of chloride of the last observation only analysis for base which was not carried out, could have determined.

Case 52 has already been mentioned. She was admitted to the hospital three times first for the usual diabetic symptoms, on both subsequent occasions because vomiting and abdominal pain had precipitated severe acidosis. In spite of the fact that she was almost comatose and the CO_2 was extremely reduced chloride had not suffered any loss. As ketosis cleared up, although she was given saline by hypodermoclysis, chloride fell as bicarbonate rose. Again the failure to determine base prevents any deductions concerning the general electrolyte level. Vomitus was not, in this case, analyzed for acid and chloride. It is, therefore impossible to say whether vomiting had resulted in loss of chloride or not. It may have been that the vomitus contained little or no chloride. The urine at the time of admission and until the diabetic toxemia had been relieved was almost free from chloride. The concentration as well as the total amount of chloride was diminished. This, as we have shown (25) is quite characteristic of the condition. At the same time the patient was extremely dehydrated. This dehydration must be brought about by several factors. Dyspnea must lead to excessive loss of fluid by evaporation. Vomiting prevents restoration of this loss and promotes further fluid depletion. The water lost by respiration carries with it no salt. The urine, as the toxemia advances contains relatively more and more water and less chloride. Unless the vomitus contains large amounts of acid or salt there must be a tendency to concentrate chloride in the body. Such a concentration may occur if the total chloride in the body remain constant or even if it diminish. This will depend entirely on the relative amounts of chloride and water excreted. Between the first two blood examinations 52 received about 7 gm of NaCl and excreted only 0.3 gm in the urine. During the same period she was given 1300 cc of fluid while only 200 cc. of urine and 200 cc of vomitus were recovered. Between the second and third examinations she received 21 gm of salt and 4000 cc of fluid and excreted 2200 cc of urine containing 6 gm of sodium chloride and vomited only

50 cc of fluid There was, then a large retention of both fluid and salt (The patient was afebrile and the respiratory ventilation diminished rapidly, so that presumably the loss of water by means of the lungs was comparatively normal) It is impossible to estimate the relative amounts of fluid and salt retained in the body because the amount of water lost by other routes and the amount of salt in the vomitus were not measured

The proportion of water and salt retained in the plasma can be calculated From the plasma protein figures it appears that the volume of the plasma at the time of the second observation was to that at the time of the first observation as $\frac{8.02}{7.45}$ or 1.08 The ratio of the chlorides

from the same plasmas was $\frac{372}{345}$ which also equals 1.08 If plasma protein may be considered a criterion of plasma volume, the actual amount of chloride in the plasma has not changed The reduction of chloride concentration is due entirely to dilution of the plasma Between the second and third examinations the proteins fell another 27 per cent, while chloride remained practically unchanged This can only mean that the actual amount of chloride in the plasma increased, but that the plasma water increased at the same rate

It appears then that blood chlorides, and probably tissue chlorides, were actually depleted at the time of admission, but that this depletion was masked by inspissation of the plasma This theory is supported by the avidity with which chloride and water were taken up by the blood and tissues during recovery

The last time this patient entered the hospital she was in even worse condition and recovery was proportionately delayed This time more studies were made although the first blood analysis, which was done in haste late at night was quite incomplete There can be no doubt this time that base was low, so low that chloride concentration must have been reduced Vomiting continued for some time This vomitus was found to contain only the most minute amounts of chloride For example the patient lost by emesis during the first 24 hours about 2200 cc of fluid, which contained altogether only about a gram of sodium chloride The sodium chloride in the urine of the same period amounted to less than 2 gm The patient had,

however, received 2000 cc of normal saline subcutaneously and a considerable but uncertain amount by rectum. Because the vomitus analyzed in the hospital was chloride free one can not argue that earlier emesis had not played a part in the reduction of the plasma salt. The relation of blood chloride to the excretion of hydrochloric acid by the stomach is still obscure. The latter may diminish as the chloride stores of the body become impoverished.

Ketosis in this case diminished but slowly in spite of the administration of large doses of insulin. Therefore, although total base returned to its normal level rapidly after the administration of large doses of salt, chloride and bicarbonate rose only gradually, yielding to the load of organic acid. On September 10, when the condition of the patient seemed to be progressing favorably a new attack of vomiting was precipitated. Within 4 hours acidosis had again developed and both bicarbonate and chloride had fallen to extremely low levels, chloride bearing the brunt of the load. Respiratory ventilation had increased to such an extent that the power of eliminating carbon dioxide must have been taxed to the utmost and chloride was forced to come to the rescue. In this instance certainly vomiting can have had little direct influence because only 0.2 gm of chloride was found in the vomitus which was recovered more than half of the total. This may be the reason that the effect of the upset on the total base was minimal. The fact that the total base concentration was relatively unimpaired enabled the blood to assume the extraordinary acid load with greater ease.

The reduction of base and chloride at the next examination may have been due to the fact that during the recovery period little attention was given to the administration of salt, while glycosuria and diuresis continued. As general improvement continued and diet and insulin were regulated all the electrolytic elements gradually returned to the normal level.

The next patient, 53, was also admitted twice. The first time he had vomited but once and from his story acidosis seemed to have developed slowly. Base figures are not available but in view of the height of total acid it can hardly have been greatly reduced. Both chloride and bicarbonate were low. With diet and insulin alone he recovered rapidly, all the acids resuming their normal values.

The second admission followed an acute attack of vomiting and abdominal pain. This had lasted only a few hours before he entered the hospital, but he was already seriously dehydrated and in a critical condition. Again base is little reduced, but the ketones have displaced both chloride and bicarbonate. There is an evident anhydremia. This time he was given immediately a hypodermoclysis of glucose and 1000 cc of normal salt. Almost no further salt was given, however, for the next two or three days. Under these circumstances, although the acidosis was overcome base and chloride remained relatively low. In this case vomiting persisted for some time after he entered the hospital, the vomitus containing small, but appreciable amounts of chloride.

The first admission of case 54 has already been mentioned. On this occasion he came in suffering from diabetic symptoms only polyuria, dehydration, loss of weight and weakness. He had ketosis with normal bicarbonate and low chloride.

The second time he was driven to the hospital by an acute respiratory infection and an active tuberculosis, in deep diabetic coma CO_2 had fallen to an almost unprecedented point while chloride had suffered little reduction. He was given 2000 cc of saline and 600 of glucose solution subcutaneously within the next twenty-four hours and as much fluid containing carbohydrate as possible by mouth. Because of his mental state the amount given by mouth was comparatively small. After this he again received little salt for the next few days. Again base fell, although chloride remained constant. In this case and the last there was apparently a certain amount of chloride reserve in the tissues that could be called upon to neutralize part of the base liberated by ketones and to maintain a constant concentration of bicarbonate in spite of the fact that the serum was diluted, but this was not enough to keep up the total salt concentration of the serum.

In contrast to these cases it may be well to turn again to case 48. This patient had a severe acidosis, but it had apparently developed gradually. At the time of admission he had moderately severe ketosis and reduction of both base and CO_2 (serum chloride was not determined). He was entirely rational and able to take a regular diet. He was given 50 gm of protein, 150 of fat and 100 of carbohydrate with

large doses of insulin at once. This insured him an adequate supply of salt. This may be the reason that during the recovery period both salt and base rose rapidly to the normal level. In fact for a while they surpassed this level.

GENERAL DISCUSSION

The general effect of ketosis on the total electrolytes of the blood is evidently somewhat variable, depending upon the severity and duration of the ketosis, and the nature of associated symptoms or pathologic conditions. If ketosis develops comparatively slowly over a long period it may attain a considerable degree of severity without producing an appreciable reduction in the concentration of bicarbonate in the blood. In these cases bicarbonate is spared at the expense of chloride, which is proportionately reduced. Occasionally the total base may increase, but this is rather unusual. More often base is diminished. In cases of severe acidosis especially if it has developed rapidly, with symptoms of grave diabetic toxemia bicarbonate has invariably been reduced. Even in these conditions, however, chloride is usually also affected. Base may be normal, but is often low and rarely, if ever, high. The ability of the organism to utilize for the neutralization of organic acid the base usually combined with chloride in addition to that derived from bicarbonate permits it to sustain a given acid load with less disturbance of the pH and a less serious demand on the respiratory mechanism.

The delivery of base by chloride may occur with extreme rapidity. In case 52 on September 10 the chloride content of serum studied only four hours after the onset of a vomiting attack which had precipitated a recurrence of ketosis and acidosis was extremely low. It is hard to escape the conclusion that in this case, at least, chloride had merely been transferred to the tissues. Certainly it was not excreted in the vomitus, and the urine of that day contained less chloride than did the urines of the preceding and following days. Unless an excessive amount was lost in the stools, which is hardly probable, the only possible inference is that the base of chloride can be rendered available for the neutralization of excess organic acids without the intervention of the kidneys, the chloride merely passing into the tissues. Once in the tissues, the chloride ions must again find base

with which to unite This may seem like robbing Peter to pay Paul In order to prevent changes in the reaction of the blood the organism immobilizes the base stores of the tissues If these stores are as limited as those of the blood such a process could only result in the impairment of the ability of the tissues to neutralize other acids It is, however, quite possible that the tissues possess reserve supplies of base in a more fixed form, in the bones, for instance, that may be mobilized to meet just such emergencies

In most of the cases with severe diabetic toxemia in this series the base of the serum was seriously diminished Often enough this depletion exhibited itself in a reduction of the concentration of base In other instances, although the concentration of base was normal, the volume of the serum had been reduced by dehydration The natural inference is that the total amount of base in circulation was diminished This inference is supported by the alterations in the electrolytes observed during recovery from toxemia In the majority of instances total base, total acid and chloride diminished during the stage of water retention that accompanied the recovery process

Although reduction of serum chloride can be effected without the intervention of excretory organs and does not necessarily imply any deficiency in the chloride content of the body as a whole one can not escape the impression that in most of these patients with severe ketosis and diabetic toxemia there is an actual depletion of the body salts and especially of chloride The process of transfer to the tissues may be only a temporary expedient for the mobilization of base to meet a sudden emergency The same forces may determine the transfer of chloride ions through the kidney into the urine in excess as the process continues and thus prevent the excessive accumulation of chloride in the tissues which would otherwise occur Loss in the feces may be increased in the same manner The fact that vomitus has been found, in a few instances, poor in chloride at a time when the serum chlorides were already low is no evidence that at an earlier period vomiting did not play an important rôle in the production of those same low serum chlorides However the chloride depletion may have been effected, circumstantial evidence of its existence is found in the persistent depression of the chloride level during recovery from ketosis If the chloride has actually escaped from the body and

is not merely segregated in the tissues it has presumably been excreted without the loss of an equivalent amount of base, the latter having been retained for the neutralization of the continuously renewed ketone acids. By only one means can the body effect such a differential excretion of as strong an acid ion as Cl , that is by the substitution of ammonia for an equivalent of fixed base.

It is not difficult to appreciate why diabetic acidosis should produce base deficiency. Every molecule of organic acid excreted must carry with it from the body into the urine a certain amount of alkali. The amount of fixed alkali thus removed is reduced to a minimum by two factors: the ability of the kidney to excrete an acid urine and to substitute ammonia for the alkaline metals. These safeguards are not, however, sufficient to spare base entirely under the strain of continued acid production. A certain amount of base is inevitably lost. Gamble, Ross and Tisdall (26) have shown in the analogous condition of starvation acidosis, that the body excretes sufficient water to maintain the concentration of base in the body at a constant level. If this explanation be accepted acidosis is the primary cause of dehydration and of salt or base depletion. J. B. S. Haldane (23) has shown that a great variety of acidifying measures lead to diuresis and water depletion.

Although the theory of Gamble and his associates explains in a satisfactory manner the phenomena of starvation acidosis, it does not entirely cover the phenomena of diabetes. The most severe stages of dehydration are undoubtedly encountered in conjunction with acidosis, but minor grades of dehydration occur at times in the absence of all evidences of acidosis and ketosis. This dehydration is presumably referable to polyuria which has in turn been ascribed to the effect of glycosuria. Unless the water loss in these cases were attended by an equivalent loss of base the concentration of salts in the body would become increased. Retention of water during recovery seems to take place even if an inadequate amount of salt is given, with the result that in many cases the concentration of base in the plasma falls as the patient first begins to improve.

Without denying that acidosis is an important agent in the production of dehydration in diabetics and with complete deference to the cogent arguments of Gamble concerning the mechanism by which acidosis produces this effect, in diabetes other forces, possibly glyco-

suria itself, may aid in provoking an excessive elimination of water. A primary loss of salt will, as Gamble has shown lead to a secondary discharge of water, it seems likely also that a primary loss of water, if it lead to true dehydration of the tissues, will be attended by an equivalent loss of salt. When the two forces are both active, as in diabetic toxemia, therefore, the body will suffer a maximum loss of both water and salts.

These observations have a certain bearing on therapy. It has already been demonstrated by Cullen and Jonas (27), Bock, Field and Adair (16) and others that insulin itself is the most efficient weapon with which to combat the acidosis of diabetes. Foster (28) has called attention to the necessity of providing with the insulin an adequate supply of carbohydrate. In this the authors heartily concur. Such carbohydrate does not only protect against the possible contingency of hypoglycemic shock. These patients have exhausted their glycogen stores. The only source from which they are able to derive the necessary antiketogenic material is the body protein. As Shaffer has shown the excess of antiketogenic over ketogenic equivalents in protein is very small. If the organism is forced to rely entirely on protein for the provision of antiketogenic materials it has no margin of safety. The failure to save 39 may have been partly due to the failure to recognize the necessity of administering glucose with insulin. On more than one occasion she recovered consciousness and her hyperpnea diminished, but the effect was only momentary. A further reason for giving carbohydrate is to diminish the destruction of protein which characterizes diabetic toxemia. It has become our custom to disregard the presence or degree of glycosuria until ketonuria and acidosis have been overcome.

The chief contraindication to the administration of carbohydrate is the fear that the persistence of glycosuria will tend to maintain diuresis and dehydration. On just this ground Bock, Field and Adair, although they employed carbohydrate freely in the treatment of diabetic acidosis, express some doubt as to the advisability of such therapy. In actual practice the persistence of glycosuria does not seem to prevent the body from retaining water. This is demonstrated by the results obtained with cases 52, 53 and 54 especially. In all these cases dilution of the blood and relief from excessive polyuria resulted long

before glucose had been eliminated from the urine. Apparently the dehydrating effect of acidosis is more powerful than that of glycosuria.

If acidosis is entirely due to the overproduction of ketone acids it can be rapidly and completely eliminated by the proper use of insulin and carbohydrate. The use of bicarbonate is not only entirely unnecessary, but even inadvisable. The combustion of ketones under the influence of insulin may in itself release so much base to combine with CO_2 as to produce an alkalosis. The administration of additional bicarbonate only heightens this effect and may precipitate edema. Whether bicarbonate is beneficial in the treatment of acidosis due to other acids or not is a point on which no one can afford to be dogmatic. Its effect is to displace from the serum an equivalent amount of chloride. It usually does, to be sure at the same time raise the pH of the blood and quiet respirations. If the non-ketogenetic organic acidosis is as we are inclined to surmise referable not to diabetes but to some associated disease its treatment can be adequately discussed only when the nature of that disease has been ascertained.

Finally, if as we have inferred chloride can aid in the maintenance of the blood reaction by yielding base for the neutralization of the ketone acids and can, by this means spare bicarbonate and relieve the respiratory mechanism, the administration of sodium chloride in conditions of acidosis would seem to be a rational procedure. If furthermore the base stores of the body are actually or relatively depleted in states of diabetic toxemia such treatment is the more imperatively indicated and should be continued during the recovery process.

The authors are quite aware that these conclusions are supported more by inference than fact. They represent the first attempt to evaluate and interpret the general electrolytic disturbances of diabetes and diabetic toxemia. Further and more extensive studies to test these hypotheses are contemplated and will be carried out as material presents itself.

SUMMARY

By a procedure outlined in a previous communication the total acid-base equilibrium of the serum or plasma in diabetes has been studied. The procedure has proved capable of detecting any considerable accumulation of ketone acids in the serum.

Ketosis of considerable severity may develop without appreciably affecting the bicarbonate of the plasma. In these cases chloride is usually found reduced and the base required for the neutralization of the organic acid is evidently derived from chloride.

In severe diabetic acidosis base for the neutralization of ketone acids may be ceded by both bicarbonate and chloride. The reduction of chloride may occur with extreme rapidity and without any appreciable augmentation of chloride excretion, indicating that the chloride ion is merely transferred to the tissues.

In profound diabetic toxemia the salt content of the blood and probably that of the tissues is seriously depleted.

The bearing of these phenomena on the treatment of diabetic toxemia, ketosis and acidosis has been discussed.

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PROTOCOLS

Case 1 A male, aged 38, laborer, entered the hospital November 22, 1922, recovering from a cold which had aggravated a mild diabetes Venous puncture with moderate stasis

Case 2 A male, aged 18, entered the hospital January 30, 1923, with a mild upper respiratory infection and glycosuria of only 2 weeks' standing (Urine had been examined at frequent intervals for 2 years because of an earlier nephritis from which he had recovered completely) His cold was subsiding and temperature normal at the time of the venipuncture

Case 3 An obese female, aged 28, who had had glycosuria without any diabetic symptoms for 2 months

Case 4 A male, aged 36, with uncomplicated diabetes of moderate severity Venipuncture at about 11 a m At this time he was receiving an adequate diet, and his urine was kept free from sugar and acetone with a single morning dose of insulin

Case 5 A stout female, aged 53, admitted to the hospital November 20, 1922, with an acute attack of recurrent arthritis with fever At the time of the venipuncture her temperature was only 99.6°F and her general condition greatly improved She had received large amounts of salicylate and bicarbonate Although sugar was found only in the first urine examination, her fasting blood sugar remained consistently elevated and she presented an excessive alimentary hyperglycemic reaction

Case 6 A moderately obese female, aged 45, admitted to the hospital November 1, 1922, for glycosuria of 4 years' duration and distinct enlargement of the liver The glycosuria responded readily to mild dietetic treatment

Case 7 A somewhat obese female, aged 56, admitted to the hospital for steadily increasing glycosuria On a liberal diet glycosuria and acetonuria disappeared rapidly

Case 8 An obese male, aged 55, admitted to the hospital February 2, 1923, with glycosuria and chronic hypertrophic spondylitis The glycosuria responded readily to mild dietetic treatment

Case 9 A moderately obese male, aged 64, admitted to the hospital April 4, 1924, with progressive dementia and mild glycosuria The latter disappeared rapidly with mild dietetic treatment Spinal fluid examination and Wassermann tests proved negative

Case 10 An obese female, aged 73, admitted to the hospital May 30, 1924, with glycosuria and severe, generalized senile eczema She presented moderate evidences of arteriosclerosis and hypertension but no signs of cardiac or renal insufficiency Her diabetes was controlled by the mildest dietary restriction

Case 11 A female, aged 27, admitted to the hospital June 10, 1924, for an uncomplicated diabetes of moderate severity which was easily controlled by dietary regulation and small doses of insulin

Case 12 A somewhat obese female, aged 43, with diabetes of 8 years' duration,

admitted to the hospital January 14, 1924, because of a recurrence of diabetic symptoms, which had set in 6 weeks earlier. No complicating condition was discovered. On the day of the blood examination glycosuria, which had proved amenable to mild dietetic treatment, suddenly recurred attended by a profuse diuresis that persisted for three days.

Case 13 A male, aged 51, admitted to the hospital May 21, 1924, with phimosis, mild balanitis, slight evidences of arteriosclerosis and mild hypertension, but no evidences of circulatory or renal failure. Glycosuria had been discovered during a routine insurance examination a few days earlier and was not attended with diabetic symptoms. At the time of the second bleeding the balanitis was cured and the diabetic condition controlled with an adequate diet without insulin.

Case 14 A female, aged 53, admitted to the hospital May 10, 1924, complaining of nervous attacks, weakness and glycosuria and recurrent attacks of gall-stone colic. X-ray revealed a large calculus in the gall bladder, and advanced chronic hypertrophic spondylitis. Her diabetes was moderately severe, but responded to diet and moderate doses of insulin.

Case 15 A female, aged 56, admitted to the hospital May 12, 1924, for attacks of weakness, associated with dyspnea, orthopnea and throbbing epigastric pain, occasional edema of the ankles, and glycosuria. Five years earlier an apoplectic stroke had resulted in temporary left sided hemiplegia, and in 1920 cardiac decompensation brought her to the hospital, where glycosuria was discovered. At the time of the venipuncture she had slight hyperpnea and orthopnea, and a few râles at the bases of both lungs, a little pitting edema of both ankles, and slight tachycardia, moderate hypertension and phenolsulfonephthalein excretion of 40 per cent.

Case 16 An obese female, aged 66, admitted to the hospital November 14, 1924, for a burning sensation and weakness of her left hand, nervousness, severe headaches, dyspnea and cardiac palpitation. Her arteries were thickened, her heart enlarged, and her blood pressure somewhat increased. At the time of the venipuncture her general condition was greatly improved.

Case 17 An obese female, aged 60, admitted to the hospital March 30, 1924, for an ulcer on the dorsum of her left foot. Glycosuria had been discovered 5 years earlier. Her heart was moderately enlarged, her systolic blood pressure 236, she had slight dyspnea, orthopnea and tachycardia and a phenolsulfonephthalein excretion of 48 per cent. There was a small superficial ulcer with little inflammatory reaction on the dorsum of the left foot. At the time of the second bleeding her general condition was greatly improved, systolic blood pressure 176, pulse and respirations normal, ulcer healed, diet adequate, no insulin and urine free from sugar.

Case 16 An obese male, aged 66, admitted to the hospital November 8, 1923, for typical diabetic symptoms of four months' duration, with the remains of a subsiding cold. The diabetes was eventually easily controlled by diet without insulin.

Case 19 A female, aged 64, admitted to the hospital February 19, 1923, because of slight cerebral hemorrhage the preceding day, with a mild diabetes discovered 14 years earlier. Her heart was moderately enlarged, her systolic blood pressure 170, her mind somewhat confused.

Case 20 A female, aged 60, admitted to the hospital September 4, 1924, for glycosuria with mild diabetic symptoms of 10 years' duration. In 1923 she had an apoplectic stroke that left her with some disability and sensory disturbances of the left hand. Her heart was moderately enlarged, her systolic blood pressure 190, but no evidences of cardiac or renal insufficiency. The glycosuria disappeared rapidly under dietetic treatment and a single small dose of insulin daily.

Case 21 A male, aged 62, admitted to the hospital May 22, 1924, with a diabetes of 4 years' standing, recently aggravated by a coryza, and jaundice which had recurred at intervals for 30 years, increasing in frequency, duration and intensity, which proved to be hemolytic in type. The diabetes was readily controlled by dietetic treatment alone. The Wassermann was negative. At the time of the second examination the patient was receiving an adequate diet and no insulin.

Case 22 A thin, but not emaciated female, aged 33, admitted to the hospital September 18, 1924, for the treatment of glycosuria and diabetic symptoms, which had developed acutely about 5 weeks earlier, shortly after the removal of an infected tooth. The glycosuria disappeared rapidly under insulin therapy and dietetic regulation.

Case 23 A thin female, aged 65, admitted to the hospital July 20, 1924, with a diabetes of 4 years' duration. Her heart was slightly enlarged, her systolic blood pressure 168 without evidences of cardiac or renal insufficiency. With insulin and dietary regulation the glycosuria soon disappeared.

Case 24 An emaciated male, aged 25, entered the hospital June 15, 1923, with diabetes discovered 14 months earlier and recently aggravated by the appearance of furunculosis. At the time of admission and venipuncture the furuncles were practically healed, he appeared somewhat dehydrated, but did not display any hyperpnea. The glycosuria proved amenable to insulin therapy.

Case 25 A poorly nourished male, aged 16, admitted to the hospital, June 9, 1923. Diabetic symptoms had come on acutely about 3 months earlier, his weight had fallen from 49.5 to 39.5 kilos. There was no evidences of any organic disease other than diabetes. Between the time of admission and the bleeding polyuria had continued and his weight had fallen another kilo.

Case 26 An emaciated male, aged 21, admitted to the hospital, January 3, 1923. He had suddenly developed acute diabetic symptoms a few weeks earlier. He appeared emaciated and dehydrated, his respirations somewhat increased in rate and depth, his urine loaded with sugar and acetone, but no complicating conditions could be discovered. Blood was withdrawn from an arm vein, with moderate stasis, at once. Glycosuria and ketonuria subsided rapidly under treatment by diet and insulin. In the course of treatment acute catarrhal jaundice, lobar pneumonia and pleurisy with effusion appeared and resulted fatally.

Case 27 A female, aged 65, admitted to the hospital March 8, 1923, with gangrene of her foot which had developed 4 or 5 weeks earlier and had aggravated a diabetes of 10 years standing which had, till then been easily controlled by dietetic treatment. For a month she refused operation and then consented only to amputation below the knee. The flaps broke down immediately. Dietetic regulation controlled the diabetes successfully except for a day after the operation when she had transient glycosuria. Her appetite failed rapidly, however, and she became stuporous. About May 5th edema appeared and, by the time of the blood examination on May 9th had become quite marked. The edema and stupor increased continuously until her death on May 18th.

Case 28 An obese female, aged 60, admitted to the hospital November 28 1924, with lacerations of the scalp and a fractured patella received in an automobile accident and a diabetes of at least 8 years' standing, which had proved amenable to moderate dietary restriction. Glycosuria disappeared rapidly with little dietary regulation as she recovered from the nervous shock and the acute trauma of the accident.

Case 29 and 45 A small, emaciated, male, aged 46, admitted to the hospital for the first time in November, 1922, with a mild acidosis. With dietetic treatment alone he was rendered aglycosuric on a diet containing only 25 gm. of carbohydrate, 50 gm. of protein and 125 of fat. Under these circumstances he developed slight, persistent, subcutaneous edema of the ankles and legs which was present at the time of the first bleeding. He remained well until a severe cold with fever and acute pain in the chest aggravated by breathing and coughing resulted in the recurrence of glycosuria and diabetic symptoms despite treatment and forced him to enter the hospital October 23. At this time he had signs of a subsiding coryza and acute fibrinous pleurisy with some evidences of acidosis. At the time of the venipuncture, the next morning before breakfast, his general condition had improved greatly.

Case 30 A male, aged 50, with moderately severe diabetes of about 3 years' duration, which had been adequately controlled by diet until aggravated by an acute upper respiratory infection, admitted to the hospital November 6, 1923, with a coryza and reddened throat, breathing quietly and apparently not much dehydrated. The first blood examination was made at once, the second the same evening after he had received 40 units of insulin.

Case 31 A small emaciated male, aged 37, admitted to the hospital March 27, 1924. For 3 years he had suffered from polyuria, polydipsia, and increasing weakness. Two weeks before admission he had developed a coryza, a week later his right ear began to discharge pus and on March 26 the left ear followed suit. At the time of the first bleeding he appeared emaciated and somewhat dehydrated, weighing only 37.7 kilos, respirations slightly exaggerated, temperature 98°F pulse 110. Both ears were discharging pus, his tonsils were inflamed. The second examination was made two days later when the patient had received adequate food, fluids and insulin and had gained 2 kilos. As the infections cleared up tolerance improved and he was discharged from the hospital, aglycosuric, with an adequate diet and a single large daily dose of insulin.

Case 32 A male, aged 57, admitted to the hospital February 20, 1925, with a long standing, mild diabetes which had been recently aggravated by an intercurrent infection. At the time of the blood examination he appeared acutely ill, his temperature 100.5°F, pulse rapid, respirations rapid and shallow, tongue dry and coated, evidences of infection of lungs, kidneys, and urinary tract and a blood culture containing pure *Staphylococcus aureus*. For the preceding 24 hours he had received little fluid.

Case 33 A female, admitted to the hospital October 25, 1922 with a fracture of the base of the skull and of the acromial end of the right clavicle, the results of a fall, unconscious. At the time of the second bleeding she had signs of consolidation of the lower lobe of the right lung, and a temperature of 104°F. It was impossible to administer adequate amounts of fluid.

Case 34 An obese female, aged 50, admitted to the hospital February 21, 1923, on the sixth day of a lobar pneumonia which had aggravated a mild diabetes of some years' duration. At the time of the first bleeding she was quite cyanotic and dyspneic, with rapid pulse, high temperature, and signs of consolidation of the whole right lung. She did not appear dehydrated and took fluids well, receiving insulin and frequent feedings of milk and cream with lactose. After the pneumonia subsided the glycosuria rapidly disappeared and she was discharged aglycosuric without insulin.

Case 35 A male, aged 65, admitted to the hospital November 28, 1924. One week earlier he was seized with pain in his left chest, bloody expectoration, and irregularity of the heart. For 2 days before admission he had become increasingly stuporous.

The night he entered the hospital he was semi-stuporous, cyanotic, with deep, rapid respirations, a totally irregular pulse, extreme cardiac enlargement, signs of consolidation over the lower lobe of the right lung, temperature of 102°F and urine containing considerable sugar and a moderate amount of acetone. He was given frequent doses of insulin, orange juice and water during the night without benefit. All specimens of urine gave heavy reactions for sugar and moderate reactions for acetone. The blood examination was made at 9 a.m. the next morning when he was almost moribund, temperature 106°F, pulse almost imperceptible. In spite of saline, glucose and insulin he died within two hours.

Case 36 A female, aged 58, with diabetes which had been well controlled for 8 or 9 years, first with diet alone and later with insulin. On September 22 anorexia and general malaise led her to omit insulin. Glycosuria increased rapidly even when insulin, without carbohydrate was resumed. She entered the hospital at 9 p.m. on September 26, emaciated, dehydrated, in deep coma, with deep accelerated, sighing respirations, a rapid, weak pulse, and profuse showers of râles over the lower lobes of both lungs, especially marked on the left side, where the breath sounds were tubular in character, and a temperature of 102°F. No urine could be obtained by catheter. Venipuncture, which was done immediately required considerable stasis. In spite of insulin, saline and glucose solutions subcutaneously she died within 4 hours.

Case 37 An obese female, aged 57. Mild glycosuria had been discovered in 1919 when she had a cerebral hemorrhage. On December 6, 1922, she developed a severe cough. Five days later she expectorated bloody sputum and became drowsy and mentally confused.

When she entered the hospital, December 13, she was semicomatose, somewhat delirious, extremely cyanotic sitting propped up in bed breathing rapidly and with difficulty, with a rattle in her throat, temperature 101°F pulse 120, systolic blood pressure 170. Over the left upper lobe and both lower lobes were dullness and tubular breathing, and coarse, bubbling rales were heard over the whole chest. Blood was examined at once and insulin administered.

The second blood examination was made the next morning. Cyanosis was less intense, tracheal rales less marked, and mental condition clearer although the whole left lung was frankly consolidated. She died suddenly a few hours later.

Case 38 An emaciated colored female, aged 70, admitted to the hospital July 11, 1924. Glycosuria with senile dementia had developed a year earlier, and symptoms of cardiac decompensation had recently appeared. She was entirely disoriented and confused, involuntary of urine and feces, breathing rapidly and noisily. Her heart was enlarged and systolic and diastolic murmurs were audible over the whole precordium. Systolic blood pressure was 180. Sibilant and sonorous rales were heard over the chest. There was well marked pitting edema of the lower extremities. A little later frank signs of pneumonia appeared and she made a rapid exodus.

Case 39 An emaciated female, aged 37, admitted to the hospital March 21, 1923. Thirty-six hours earlier she suddenly developed acute abdominal pain, nausea and vomiting. By the next morning she was weak and stuporous and within 24 hours had sunk into deep coma.

She was admitted in deep coma, breathing rapidly and extremely deeply, emaciated and dehydrated, with systolic blood pressure only 88. Blood was examined at once. After insulin and a hypodermoclysis of saline she recovered consciousness, only to relapse rapidly in spite of further treatment. Vomiting continued. The urine output was scanty and further injections of saline resulted only in the production of subcutaneous edema. No carbohydrate was injected. She died within two days without the appearance of further symptoms to explain the origin of the acute illness. No history of diabetes preceding the present illness could be obtained.

Case 40 An emaciated female, aged 64, admitted to the hospital October 16, 1924. Mild diabetes had been recognized for a year previous. For 4 days before admission she was unable to move her bowels in spite of numerous cathartics and had taken almost no food nor fluids, but had not vomited.

On admission, when the blood examination was made, she appeared weak, emaciated and dehydrated. Her abdomen distended with gas, her rectum filled with impacted feces. Cleaning out the rectum thoroughly with enemata, regulation of diet, small doses of insulin and treatment of hemorrhoids resulted in alleviation of all symptoms.

Case 41 An obese female, aged 61, admitted to the hospital September 6, 1924, for treatment of a gangrenous toe which had aggravated a mild diabetes of 10 years' standing. The diabetes was readily controlled by diet and insulin and the toe healed without surgical interference. While she was in the hospital she had an attack of biliary colic and jaundice which subsided spontaneously and for which she refused operation. She also had an enormous ovarian cyst and moderate hypertension with some evidences of beginning heart failure.

October 22 she developed dull pain in the abdomen, nausea, and vomiting, and jaundice. When the blood was examined, October 25, she was deeply jaundiced, acutely ill and dehydrated with rapid, irregular pulse, but normal respirations, and enlarged liver and tenderness in the right upper quadrant. October 28 the gall bladder was drained. The second blood examination was made just after the operation. Intractable distention developed and she died October 31.

Case 42 A male, aged 46, admitted to the hospital February 4, 1925, with an acute appendicitis with abscess. Extensive necrosis of the cecum necessitated resection and end to side anastomosis of the ileum and cecum. Three days later persistent vomiting and abdominal distention appeared. February 8 urine contained no sugar, but considerable acetone. His blood sugar at 11 a m. was 366 mg per cent. He was given 1500 cc saline subcutaneously at noon and at 5 30 p m. 500 cc of 5 per cent glucose and 400 cc of saline with 20 units of insulin. Cecostomy was performed about 2 hours later. The blood was examined about 2 hours after the operation, when his temperature was 102°F. He recovered after a protracted illness without further recurrence of glycosuria or hyperglycemia.

Case 43 An emaciated male, aged 18, admitted to the hospital September 17, 1924. For 3 years he had had a diabetes, controlled with difficulty by diet and insulin. On September 14 he became soaked and fatigued, exhausted his supply of insulin, and took one or two large meals. The next day nausea and vomiting set in and he continued without insulin. By night he was almost comatose, overventilating and excreting large amounts of glucose and acetone. He received only broth and 60 units of insulin in the next 24 hours, at the end of which time he was brought to the hospital. On admission, when the first venipuncture was made he seemed tired and drowsy, but mentally clear, extremely dehydrated, breathing quietly, with rapid pulse. Vomiting had ceased. During the course of the night he was given 50 units of insulin and large amounts of fluid containing carbohydrate. The next morning he was able to resume his former diet. The second blood examination was made before breakfast 5 days later. Meanwhile he had gained more than 3 kilos of weight and his urine had been free from sugar and acetone for 24 hours.

Case 44 An undernourished male, aged 26, admitted to the hospital February 12, 1923, for treatment of diabetes which had developed acutely 3 years before and recently been aggravated by a coryza. At the time of admission when the first blood was taken he appeared dehydrated and presented definite hyperpnea. He weighed only 47.7 kilos. At the time of the second examination 10 days later his weight had increased to 64.3 kilos although he was receiving only 1340 calo-

ries a day and he had marked, generalized, subcutaneous edema, ascites and double hydrothorax. When his diet was increased to 1450 calories daily his edema rapidly disappeared without other treatment. With dietary regulation and insulin regained weight and strength and was able to resume his normal occupation.

On August 25, 1923 he was again admitted with all the evidences of acidosis and dehydration, precipitated by a corvza. The third blood was taken as soon as he entered the hospital. He recovered rapidly and his carbohydrate tolerance returned to its former level.

The last blood was examined when he called to report progress. He had been continually aglycosuric, had regained his normal weight, and his carbohydrate tolerance had not diminished.

Case 45 (See no. 29)

Case 46 A male, aged 23, admitted to the hospital August 12, 1924. Diabetic symptoms had begun suddenly 8 months earlier. He first came to the outpatient department January 6, 1924, weighing 78.7 kilos, large and well-developed, not obese, without evidences of organic disease other than diabetes. With moderate dietary restriction glycosuria and diabetic symptoms disappeared, but again recurred in May.

When he entered the hospital August 12 he weighed only 63.2 kilos and appeared weak and somewhat dehydrated. His blood sugar before breakfast was 210 mg per cent and his urine contained large amounts of sugar and acetone. On a diet of 65 gm of protein, 150 of fat and 100 carbohydrate and 55 units of insulin, as glycosuria and ketonuria diminished his weight increased rapidly. On August 22 he presented well marked edema of the ankles and the next day, when the first blood was taken, weighed 67.3 kilos. Without change in treatment, but with a spontaneous increase of carbohydrate tolerance the weight began to fall the next day. He was discharged September 2, weighing 64.1 kilos, aglycosuric, free from edema, on a diet containing protein 65 gm., fat 200, carbohydrate 100, 2537 calories, taking only a single dose of 15 units of insulin daily. The second blood was taken August 31, when the edema had disappeared.

Case 47 A very small emaciated male, aged 27, admitted to the hospital March 4, 1925. Severe diabetic symptoms had suddenly developed 4 months before this after a severe cold, and were again aggravated 2 months later by an acute otitis media. When he entered the hospital he weighed only 30.6 kilos, 20 kilos less than he had weighed before the onset of the diabetic symptoms, and had extreme polyuria. He appeared emaciated and dehydrated, his respirations were somewhat increased, purulent discharge issued from his left ear and his tonsils were slightly inflamed. The first blood was taken at once.

On a diet of 50 gm of protein, 150 of fat and 100 of carbohydrate and increasing doses of insulin glycosuria, acetonuria and polyuria rapidly disappeared and his weight increased, reaching a maximum of 37.2 kilos on March 17. From this point on it again diminished rapidly, the edema disappearing as his general condition and tolerance improved. At the time of the second blood study the acidosis had disappeared, at the time of the third edema was quite evident (weight 36

kilos), at the time of the fourth edema was no longer evident (weight 34 kilos). At the time of the last bleeding he was aglycosuric on a diet containing 60 gm of protein, 150 of fat and 110 of carbohydrate with 30 units of insulin daily and weighed 34.5 kilos.

Case 48 A female, aged 47, admitted to the hospital December 11, 1922. One year earlier, when she had furunculosis, otitis media and pyuria, glycosuria had been discovered, which cleared up when the furunculosis disappeared. Two weeks before admission she developed a cold and cough, which increased in severity. December 7th persistent, distressing hiccough and vomiting appeared, followed by increasing drowsiness and dyspnea, for which large amounts of sodium bicarbonate and fruit juices were given.

When she entered the hospital and the first blood was drawn she was quite stuporous, but restless, slightly cyanotic, breathing slowly and superficially. Her heart was enlarged, pulse 101, systolic blood pressure 180. Harsh breathing and scattered râles were heard over both sides of the chest. There was slight subcutaneous edema of the ankles and shins. The urine contained large amounts of pus, and considerable sugar and acetone.

Thirty units of insulin were given in the course of the night and, by the next morning, her urine contained neither sugar nor acetone, she was mentally clearer, and the râles in the chest were less profuse. Hiccough and vomiting persisted and interfered seriously with the administration of fluid.

The following day, when the second blood was taken improvement was more marked, but the edema had increased and vomiting continued. The vomitus contained no free HCl and almost no combined acid. In the course of the day she received 1000 cc of normal saline solution by hypodermoclysis. Vomiting and edema disappeared rapidly and she improved steadily from this time on.

The third blood was taken January 11, when she was receiving a diet containing 50 gm of protein, 100 of fat and 75 of carbohydrate, without insulin. Her urine still contained pus and a faint trace of acetone, her lungs were clear and there were no signs of edema.

Case 49 A female, aged 57, admitted to the hospital August 16, 1923 with gangrene of the foot which had aggravated a mild diabetes of 7 years standing.

When she entered the hospital she was semi-stuporous, breathing rapidly and rather deeply, and appeared somewhat dehydrated. The right little toe and the adjacent portion of the dorsum of the foot were black and gangrenous and there were evidences of spreading cellulitis extending from the gangrenous area. Operation was refused by the family. In spite of increasing doses of insulin and large amounts of fluid containing carbohydrate it was impossible to control the diabetes continuously in the face of an advancing infection and constantly increasing temperature and toxemia, as is evidenced by the result of the first six blood examinations, the first of which was made as soon as she entered the hospital and the other five before breakfast on the days indicated in the table.

The leg was amputated above the knee August 20, shortly after the sixth examination, and the seventh was made before breakfast the next morning. She

improved for the first few days after the operation, but the flaps then broke down and she sank gradually. In spite of the fact that the diabetes was adequately controlled by diet and insulin, she died about a month later.

Case 50 A female, aged 56, admitted to the hospital January 28, 1924. Five years earlier she had a sudden attack of unconsciousness lasting several days during which glycosuria was discovered. After that she remained free from symptoms with moderate dietary regulation although she showed a slight, persistent glycosuria. Forty-eight hours before she entered the hospital she seemed confused when she awoke in the morning. She became unconscious in a few hours and shortly after this hyperpnea and acetonuria appeared.

The first specimen of blood was taken just after she entered the hospital, when she was in deep coma, breathing very deeply, 26 to the minute, and markedly dehydrated. Her temperature was 101°F, pulse 108, systolic blood pressure 140. Over the base of the right lung there was dullness, with diminished breath sounds and râles. Her blood count showed 6.3 million red blood cells and 27,000 leucocytes with 91 per cent of polymorphonuclear neutrophils. Urine, obtained by catheter, contained a very faint trace of albumin, many hyaline and granular casts and large amounts of sugar and acetone.

She was at once given hypodermoclysis containing 500 cc. of 5 per cent glucose solution and 1000 cc. of normal saline and 12 hours later another of 500 cc. of glucose and 300 cc. of saline. She also received orange juice sweetened with sugar, by mouth, and 70 units of insulin in frequent divided doses. The next morning when the second examination of the blood was made her respirations were quiet, her pulse rate was normal and her urine contained considerable sugar, but only a trace of acetone. Her temperature was still 100°F and she was still in deep coma.

In the next 24 hours she received 5600 cc. of fluid and large amounts of sugar. This included a hypodermoclysis of 500 cc. of 5 per cent glucose solution and 1000 cc. of normal saline. When the third blood was taken on the 30th, her mental state was somewhat clearer, but she remained quite stuporous, unable to respond to questions and her right hand seemed distinctly weak. Her urine contained little sugar, but no acetone.

After this she improved rapidly. On February 1 she had roused from her stupor, but presented a definite motor aphasia, slight dysphagia, and weakness of the right arm and the right side of her face.

At the time the fourth blood was taken she was receiving a diet containing 50 gm. of protein, 100 of fat and 100 of carbohydrate, and 60 units of insulin daily. The aphasia persisted, her pulse was quite irregular, with frequent extrasystoles, and the electrocardiograph subsequently revealed evidences of coronary disease. The signs in the lungs remained unchanged, temperature 100.8°F.

The patient was discharged from the hospital some weeks later, without glycosuria, receiving an adequate diet and only 15 units of insulin daily. The aphasia did not clear up.

Case 51 A poorly nourished female, aged 50 first admitted to the hospital

in July, 1923, with a moderately severe diabetes which was controlled by a diet containing 50 gm of protein, 175 of fat and 75 of carbohydrate, with 15 units of insulin daily. She did not adhere strictly to this diet, glycosuria recurred and increased in spite of increasing doses of insulin. In September polyuria, polydypsia and weakness developed and one day, while shopping, she fainted. On November 15 she had a sudden attack of abdominal pain and vomited. The pain and vomiting persisted and she became increasingly drowsy. She was brought to the hospital November 19, mentally clear, but extremely drowsy and exhausted, markedly dehydrated, respirations deep and somewhat rapid, pulse rapid, but regular. There was some pain and tenderness in the epigastrium, but no other signs of organic disease. The blood count showed 6,100,000 red blood cells and 13,200 leucocytes with 76 per cent polymorphonuclear neutrophils. Her urine contained 7.6 gm per liter of sugar, large amounts of acetone and diacetic acid, a trace of albumin and a few hyaline casts. The first blood was taken at once.

The patient was immediately given insulin and a hypodermoclysis of 1000 cc of normal saline solution. At the same time dilute sugar solution was administered by mouth with frequent doses of insulin. By the next morning, when the second sample of blood was examined, her mental condition was clearer and she was breathing more quietly. Her urine contained about 20 gm of glucose per liter, but considerably less acetone.

The administration of large amounts of fluid, carbohydrate and insulin was continued and by the morning of November 21, when the third blood was taken, respirations were normal and the urine free from acetone, although it still contained 4 per cent of sugar. She still vomited small amounts occasionally.

After this she improved steadily. Vomiting and other symptoms disappeared and she was able to resume a mixed solid diet. When the fourth blood was taken, November 27, she was receiving a diet containing 55 gm of protein, 200 of fat and 70 of carbohydrate, with 120 units of insulin daily, and her urine showed no acetone nor diacetic acid and only an occasional trace of sugar. At this time she weighed 56.7 kilos.

When the fifth blood was taken she was receiving the same diet, but only 70 units of insulin.

After she left the hospital she was forced to increase her insulin to 130 units daily, but on this régime and in spite of frequent dietary lapses she remained aglycosuric and free from symptoms. In July she tried to reduce the insulin but glycosuria and weakness at once developed. About the same time she developed a furuncle on her leg which temporarily reduced her tolerance further. In the middle of August she again cut down her dosage of insulin against advice. Two weeks later she began to suffer from weakness and increasingly frequent attacks of dizziness, cardiac palpitation and breathlessness, occasionally resulting in syncope. On September 5, after one of these attacks she was suddenly seized by abdominal pain and a little later vomited. From that time until she entered the hospital at 10 p m the next day, September 6, she vomited continuously.

Blood was taken as soon as she entered the hospital. She was drowsy and complained of continuous nausea, extreme thirst and slight abdominal pain. She was extremely dehydrated, her respirations were somewhat rapid, very deep, and interrupted at frequent intervals by retching and vomiting. Her pulse was rapid and quite irregular, apparently because of the occurrence of frequent extrasystoles; her heart sounds weak, distant and extremely irregular. Her lungs were clear, the abdomen everywhere tender, but not resistant. Her urine contained 2.1 per cent of sugar only, 0.02 per cent of sodium chloride, and gave a heavy reaction for acetone. She received at once 1000 cc. of normal saline and 500 cc. of 5 per cent glucose solution subcutaneously together with 40 units of insulin and was allowed water and sweetened orange juice by mouth.

At 1 a.m. September 7, her condition was little improved so another 40 units of insulin was given, followed by two 20 unit doses at 3 and at 7 a.m. The second blood was taken at 9 a.m. By this time nausea and abdominal pain were less severe, her pulse was slower and more regular, the respirations somewhat quieter. She complained of severe headache and vomited at intervals. Her urine contained 2.5 per cent of sugar, 0.02 per cent of NaCl and much acetone. During the day as vomiting continued, another hypodermoclysis of 1000 cc. of saline and 500 cc. of 5 per cent glucose was given and peroral fluids were limited to 50 cc. of water every half hour. This checked the vomiting and by midnight she seemed greatly improved.

The third blood was taken at 9 a.m., September 8. At this time, although her urine still contained 2.3 per cent of sugar and gave a heavy reaction for acetone, her respirations were normal, pulse regular, and vomiting had ceased. Improvement continued throughout the day and it proved possible to increase the diet somewhat. In spite of large doses of insulin sugar disappeared from the urine at intervals only.

Early in the morning of September 10, she got out of bed without permission and at once became dizzy and vomited. Cardiac palpitation and abdominal cramps soon developed, she vomited once or twice more and within an hour presented air hunger and all the other evidences of severe acidosis. By 10 a.m., 5 hours after the onset of the attack, when the fourth blood was taken she was again overventilating, her pulse was rapid and weak, she appeared dehydrated and complained of extreme thirst. Diet was stopped, she was given an enema and allowed only small doses of orange juice and water by mouth. Vomiting ceased rapidly and by the next morning she was greatly improved and able to resume her diet in frequent, small feedings.

On the night of September 11, she had an attack of cardiac palpitation and dyspnea. At 5 a.m. the next day, September 12, she was awakened by severe abdominal cramps and a little later vomited a large amount of yellowish fluid. The symptoms this time subsided rapidly. By 9 a.m., the pain had subsided. Although she had not vomited again she exhibited evident hyperpnea. Improvement now proceeded rapidly. On September 13 the urine finally became free from sugar and acetone. During the night she had a slight attack of palpitation

which caused the appearance of considerable sugar and acetone in the next specimen of urine voided. Otherwise recovery was uninterrupted.

The three last examinations of blood were made before breakfast on the days indicated in table 2 and mark the progress of convalescence. She was finally discharged from the hospital October 7, aglycosuric, receiving a diet containing 60 gm of protein, 200 of fat and 100 of carbohydrate and 90 units of insulin daily.

An attempt is made in table 3 to present the metabolism data in abstract form.

Case 52 An emaciated male, aged 36, first admitted to the hospital February 8, 1924. Three months earlier he had been suddenly seized with sharp, cramp-like pains in the upper part of the abdomen, which lasted about 24 hours. Immediately after this he developed extreme thirst and polyuria, which lasted about a week, gradually diminishing in intensity, attended by rapid loss of weight and strength. (On February 5 he vomited again once and had a recurrence of diabetic symptoms.) During the first week of his illness he lost 14 kilos and subsequently had lost another 10.

When he entered the hospital he complained especially of anorexia and extreme weakness. He appeared seriously ill, emaciated and dehydrated, with moderate hyperpnea. Except for the presence of pyorrhea and several bad teeth, examination revealed no evidences of any organic disease other than diabetes. His urine contained large amounts of sugar and acetone. The first blood was taken the next morning when his condition was practically unchanged and he had received nothing but non-nutritive fluids. He was given orange juice and water with large doses of insulin at frequent intervals throughout the day and, by the next morning, February 10, when the second blood was taken, his condition was greatly improved, his urine was free from sugar and gave only a moderate reaction for acetone and he felt able to take a regular diet. He was accordingly ordered 50 gm of protein, 150 of fat and 100 of carbohydrate daily and insulin was adjusted as rapidly as possible. Under these circumstances his weight increased at the rate of a pound a day for two weeks, at the end of which time he presented slight edema of the ankles. The diet was subsequently increased and insulin dosage reduced as he improved and he was finally discharged from the hospital on March 7, aglycosuric, receiving a diet which contained 50 gm of protein, 200 of fat and 100 of carbohydrate, without insulin. The third blood was taken when he was in excellent condition, free from edema and glycosuria.

After he left the hospital he was careless about his diet and habits. He also had a few minor disorders of digestion. His carbohydrate tolerance diminished and he was forced to first resume and then to increase insulin therapy until he was taking 40 units a day.

November 9 he did not feel well and was unable to eat any supper. At 3 a m the next morning he awoke nauseated and vomited. Almost immediately he developed severe epigastric pain. From that time until he entered the hospital, at 2 p m the pain persisted and he vomited every time he tried to take anything by mouth. For 3 days preceding this attack he had been eating and drinking carelessly and had been without insulin.

Date	Intake						Urino			Vomit				Treatment
	Protein	Fat	Carbohydrate	Fluids	Cl as NaCl		Volume	Sugar	Cl as NaCl	Volume	Sugar	Cl as NaCl	Insulin	
	gm	gm	gm	cc	gm		cc	gm	gm	cc	gm	gm	units	
September 6*	0	0	25+	1,500+	9.0		440	12	0.1	710	6.2	0.1	90	1000 cc of saline and 500 cc 5 per cent glucose were given subcutaneously. An uncertain amount of water and sweetened orange juice were taken by mouth.
September 7	0	0	70+	6,500	6.3	1,750	15	3.5	3.1	310	2.8	0.6	140	700 cc of saline and 500 cc 5 per cent glucose subcutaneously and 15 grams of sugar as orange juice by mouth. Glucose was also given by rectum, but only a small and uncertain amount was retained.
September 8	30	90	100	2,500	2†	2,370	83	9.6	0	0	0	0	90	Diet of orange juice, milk, cream, lactose and ten and toast.
September 9	48	100	100	1,200	2†	1,060	131	7.1	0	0	0	0	100	Diet of same character, but more liberal.
September 10	0	0	62	2,500	0	2,250	57	1.5	1,000†	6†	0	1†	120	Only sweetened orange juice and water given.
September 11	35	100	75	2,100	2†	2,220	71	5.0	0	0	0	0	110	Diet similar to those of September 8 and 9.
September 12	22	78	19	1,900	2†	1,540	18	3.8	180	0	0	2	130	Similar diet, somewhat reduced in amount because patient vomited once.
September 13	15	100	75	3,300	2†	2,910	54	2.9	0	0	0	0	90	Diet was gradually altered in quality to conform more nearly to the normal character by the introduction of small amounts of meat and cereal.
September 14	15	100	75	3,800	2†	1,950	21	2.2	0	0	0	0	110	
September 15	15	100	75	3,000	2†	1,400	32	1.7	0	0	0	0	110	
September 16	15	100	75	1,300	2†	1,500	17	3.9	0	0	0	0	80	

* 1 from time of admission, 10 p.m. September 6, to 7 a.m. the next morning.

† Only about one-half the vomitus, 510 cc, was actually recovered and analyzed.

sugar

‡ The chloride content of these diets was uncertain. From the nature of the food stuffs employed they must have been comparatively low in chloride.

His contained 0.2 grams NaCl and 3 grams of

When he entered the hospital and the first blood was taken he appeared desperately ill, semi-stuporous, emaciated and dehydrated, with extreme hyperpnea, rapid, weak pulse, a temperature of 99°F , systolic blood pressure 112. The abdomen was everywhere rigid, and there was an area of marked tenderness just to the left of the naval. His blood count showed 5.6 million red blood cells and 30,000 leucocytes with 86 per cent polymorphonuclear neutrophils. The urine contained a moderate amount of albumin, many granular casts, 3.5 per cent of sugar and considerable acetone.

He was given immediately 1000 cc of normal saline and 500 cc of 5 per cent glucose solution by hypodermoclysis with 40 units of insulin. Urine was collected as frequently as possible, and, as glycosuria persisted, 2 additional 20 unit doses of insulin were given in the course of the afternoon. By 6 p.m. the abdominal pain and tenderness had disappeared, but he was still partially stuporous and his respirations were still greatly exaggerated. He had vomited coffee-ground material twice. From this time on until the next morning he received 20 units of insulin every two hours and 5 gm of carbohydrate in the form of sweetened orange juice and water every half hour. The second blood examination was made at 10 p.m. that night and the third at 8.30 a.m. the next morning. Urinary sugar had not diminished, but ketonuria had practically disappeared by this time.

From this time on his condition improved steadily. Vomiting did not recur. He was able to resume his diet and glycosuria disappeared. He was finally discharged from the hospital on December 4, aglycosuric, receiving a diet containing protein 60 gm, fat 200 gm, and carbohydrate 100 gm with 60 units of insulin daily.

Up to the time of the third examination, before breakfast on November 13, he had received only sweetened orange juice, two slices of bread, a small amount of milk and cream and one cup of soup. The total salt content of the diet must, therefore, have been minimal.

Case 53 A male, aged 33, was admitted to the hospital March 26, 1924, for the treatment of glycosuria and diabetic symptoms which had developed quite suddenly a few weeks earlier. (The patient had a bilateral herniotomy performed in this hospital in November, 1923. At that time urine both before and after operation was free from sugar.) He appeared comparatively well nourished, but somewhat dehydrated, and complained of continuous thirst and polyuria. Examinations, including x-ray of the chest and examination of the sputum, revealed no evidences of any organic disease other than diabetes. His urine contained large amounts of sugar and acetone. The first blood was taken the next morning, when his condition was practically unchanged. The second was taken two days later. By this time, with insulin and dietetic treatment, his general condition had improved greatly, he had gained 5 pounds in weight and his urine contained only a trace of sugar and a moderate amount of acetone. Improvement continued uninterrupted and he was discharged on April 18, aglycosuric, receiving a diet containing 70 gm of protein, 200 of fat and 100 of carbohydrate, with 10 units of insulin daily.

After he left the hospital he continued his treatment with excellent results for some time. About the first of January, 1925, he stopped taking insulin. Polyuria and polydipsia returned and began to increase steadily in intensity. In a few weeks he lost 27 kilos of weight. In spite of this he became more and more careless of his diet and continued without insulin. About the middle of February he developed a productive cough which grew continuously more severe. March 18 weakness and chill sensations appeared and two days later substernal pain. The next day he had a distinct chill, vomited several times, and gradually sank into a stupor. He was brought to the hospital in deep coma March 22, extremely dehydrated, with maximal respirations, cold and somewhat cyanotic, with subnormal temperature, rapid, weak pulse, a systolic blood pressure of only 80, his throat inflamed and edematous, and dulness, suppressed breath sounds and showers of râles over the whole right chest below the third rib. His blood count showed 5.2 million red blood cells and 11,700 leucocytes with 83 per cent polymorphonuclear leucocytes, tubercle bacilli were found in his sputum and subsequent x-ray examination revealed an extensive lesion of the right lung. His urine contained large amounts of sugar and acetone, a trace of albumin and many granular casts.

The first blood examination was made at once and he was immediately given 30 units of insulin and 200 cc. of orange juice followed shortly by a hypodermoclysis of 1000 cc. of normal saline and 500 cc. of 5 per cent glucose solution and another 30 units of insulin. During the remainder of the night he received 80 units of insulin in addition and 120 gm. of carbohydrate in the form of sweetened orange juice.

The next morning he was given another hypodermoclysis of 1000 cc. of saline and 500 of glucose solution and in the afternoon an additional 1000 cc. of 5 per cent glucose. By 2 p.m. he began to respond to questions. This day, March 23, besides the fluids administered subcutaneously, he received about 200 gm. of carbohydrate and a minimal amount of protein and fat in the form of orange juice, lactose, milk and cream. He was given 70 units of insulin in the 24 hours.

The next morning, March 24, when the second blood was taken he had improved greatly. His urine contained only a moderate amount of sugar and no acetone. His respirations were quiet and normal and he was more rational. His diet was varied somewhat by the addition of eggs and he was given less insulin. By the following morning he was quite rational and able to take a diet containing 60 gm. of protein, 150 of fat and 100 of carbohydrate.

The morning of the 27th when the last blood was examined, he was receiving this diet. His urine at the time of the venipuncture contained considerable sugar but no acetone.

His temperature, which had been subnormal when he entered the hospital, rose to 100°F, on the second day and remained somewhat elevated for about a week, when it fell to normal. The general condition of the patient and his tolerance for carbohydrate improved so much that he was able at one time on a diet containing protein 75 gm., fat 250 gm. and carbohydrate 125 to reduce insulin to 10 units a day without the appearance of glycosuria.

TOTAL ACID-BASE EQUILIBRIUM OF PLASMA IN HEALTH AND DISEASE

VII FACTORS CAUSING ACIDOSIS IN CHRONIC NEPHRITIS A PRELIMINARY REPORT

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(Received for publication August 6, 1925)

Throughout this paper the term acidosis is used in the sense in which it was employed by Van Slyke and Cullen (1) to define "a condition in which the bicarbonate of the blood is reduced below the normal level." They have pointed out that the bicarbonate represents the excess of base which is left after all of the non-volatile acids have been neutralized and that this is the base available for the immediate neutralization of any further increments of acids.

Acidosis is an almost constant feature of the later stages of chronic interstitial and arteriosclerotic nephritis. In a series of such cases the inorganic constituents of the serum have been determined in order to obtain some indication of the cause of the acidosis. Studies of other types of kidney insufficiency have not been included in this report.

EXPERIMENTAL PROCEDURE

In most cases arterial blood was obtained in the morning before breakfast and the plasma or serum separated at once without exposure to air. In a few cases plasma obtained from venous blood collected without stasis was used. In a few other cases venous blood was saturated at 38°C with CO₂ at 40 mm pressure and the plasma then obtained. The determinations in earlier studies were made on plasma from oxalated blood while later ones were on serum. Blood in these later studies was defibrinated by continuously inverting and reinverting the sampling tube containing the blood and a small amount of mercury. With care serum can be obtained in this way without hemolysis. Further details of the technique used in handling blood have been described in previous articles (2).

Total base was determined by Cullen and Robinson's¹ adaptation of Fiske's urine method. Chloride was determined by Austin and Van Slyke's (3) method in earlier studies and Van Slyke's method (4) in later studies, CO₂ by Van Slyke and Stadie's method (5) and phosphate by Briggs' (6) or Benedict and Theis' (7) modification of Bell and Doisy's method. The protein was calculated from the nitrogen determination by the ordinary Kjeldahl method corrected for the non-protein nitrogen. The non-protein nitrogen as determined on whole blood was used for this correction instead of actual plasma figures. The error incurred by this procedure is slight.

Base combined with protein at any given pH was calculated by the formula devised by Van Slyke, Wu and McLean (8) from experimental data. In calculating the base combined with phosphate it was assumed that in each sample of plasma or serum mono-basic and di-basic phosphate were present in the ratio 1:4. Theoretically this ratio varies somewhat according to the pH. The error involved in neglecting these variations is slight. In an extreme case with 15 mg of inorganic phosphate per 100 cc of serum and a pH of 7.00 the base combined with phosphate calculated by this method would be one millimol too high. In the cases where pH was not determined and the CO₂ tension was not known, a pH of 7.35 was arbitrarily assumed in calculating the dissolved CO₂ and the base combined with protein.

EXPERIMENTAL RESULTS

Besides the reduction of bicarbonate one frequently finds a reduction of plasma chloride. The frequency and degree of this reduction is illustrated in table 1. (The values throughout these tables are expressed in mM of base-combining capacity). In normal subjects the chloride fell between 100 and 110 mM, in 77 determinations in nephritis much greater variations were encountered. In over one-half of the determinations chloride was distinctly below normal, in 5 above normal. The magnitude of the chloride deficit frequently exceeded the total capacity of bicarbonate.

Table 2 shows the variation in the base bound by bicarbonate, chloride and protein combined, the 3 most significant acids of normal plasma. Normal subjects varied between 135 and 145 mM. Low values were found in 66 out of 77 determinations on cases of nephritis and in more than one-half the reduction was considerable.

Retention of phosphate has commonly been considered an important factor in the production of acidosis in chronic nephritis. In table 3 are recorded 46 determinations which also included the base bound with phosphate. High phosphate was found in most

¹ Personal communication

of these cases but the phosphate increases were quite insufficient to bring the sums of the acids to normal. In 35 out of 46 determinations base combined with bicarbonate, chloride, protein and phosphate was definitely reduced.

Total base as well as the acids already enumerated have been determined 25 times in nephritis. Serum being practically neutral, the

TABLE 1
Chloride variation in chronic nephritis

	NUMBER OF DETERMINATIONS	NUMBER OF CASES	LIMITS OF VARIATION mM
Normal			100.0-110.0
Nephritis, total	77	28	67.3-127.1
Nephritis	8	5	67.3-80.0
Nephritis	20	9	80.0-90.0
Nephritis	18	11	90.0-100.0
Nephritis	26	13	100.0-110.0
Nephritis	5	4	110.0-127.1

TABLE 2
Variation of base combined with bicarbonate, chloride and protein

	NUMBER OF DETERMINATIONS	NUMBER OF CASES	LIMITS OF VARIATION mM
Normal			135.0-145.0
Nephritis, normal	77	28	91.0-150.1
Nephritis	6	5	91.0-105.0
Nephritis	12	8	105.0-115.0
Nephritis	23	11	115.0-125.0
Nephritis	23	13	125.0-135.0
Nephritis	11	8	135.0-145.0
Nephritis	2	2	145.0-150.1

sum of the total acids must equal the base. Base may be considered a measure of the total electrolyte concentration. In normal subjects base has varied between 150 and 165 mM. In the 25 determinations on cases of nephritis it has been definitely reduced in 12 and above normal in three (table 4). A reduction of the total electrolyte level is therefore one factor in the mechanism producing acidosis and low chloride in nephritis.

Gram (9) found the freezing point of sera from cases with uremia lower than normal, indicating to him an increase in osmotic pressure. Associated with this he found a decrease of chloride and electrical conductivity. He suggested that there was a decrease of electrolytes in the serum to compensate for the osmotic effect of the increase of nitrogen waste products. In this series no clear relationship can be established between the amount of base and the level of non-protein nitrogen.

The question arises to what extent other undetermined acids may have replaced bicarbonate and chloride from combination with base. This is indicated in table 5 which is a study of the difference between the total acids determined and the total base. This difference in

TABLE 3
Variation of base combined with bicarbonate, chloride, protein and phosphates

	NUMBER OF DETERMINATIONS	NUMBER OF CASES	LIMITS OF VARIATION
			<i>mM</i>
Normal			136 0-148 0
Nephritis, total	46	16	100 7-152 6
Nephritis	3	1	100 0-115 0
Nephritis	11	6	115 0-125 0
Nephritis	21	10	125 0-135 0
Nephritis	11	5	135 0-152 6

normal subjects has always been less than 20 mM. Out of 25 determinations in this type of nephritis it was greater than 20 in eighteen. In this we have an additional and important factor producing the low bicarbonate and chloride. If, as Denis (10) has suggested, high sulfate is of common occurrence among these cases, part of the augmented acid deficit presumably represents sulfate. This can hardly account for the total and one is forced to the conclusion that organic acids must also play a part.

It seems clear therefore that more than one factor is active in the mechanism producing acidosis in chronic nephritis. Retention of phosphate is almost invariably present in the later stages of disease. Increase in serum phosphate plays a significant rôle in reducing the base available for bicarbonate but in the majority of cases this is not

the most important factor The diminution of total base is frequently quite striking and in addition there is evidence of a marked increase in organic acids

Marrack (11) has made a study of the inorganic constituents of sera from cases of chronic nephritis He determined only the bicarbonate chloride, phosphate and sodium but this was sufficient to indicate

TABLE 4
Variation of total base in chronic nephritis

	NUMBER OF DETERMINATIONS	NUMBER OF CASES	LIMITS OF VARIATION
			mM
Normal			150 0-165 0
Nephritis, total	25	13	135 1-199 2
Nephritis	5	2	135 1-145 0
Nephritis	7	6	145 0-150 0
Nephritis	10	6	150 0-165 0
Nephritis	3	2	165 0-199 2

TABLE 5
Variation of undetermined acids in chronic nephritis

	NUMBER OF DETERMINATIONS	NUMBER OF CASES	LIMITS OF VARIATION
			mM
Normal			Less than 20
Nephritis, total	25	13	46 6- 6 1
Nephritis	6	6	46 6- 30 0
Nephritis	7	7	30 0- 25 0
Nephritis	5	5	25 0- 20 0
Nephritis	7	6	Less than 20

changes similar to those described here He believed the most important factor causing the acidosis was an increase of undetermined anions In a few cases low values for sodium were found Fiegl (12) and later Denis and Hobson (13) made studies of the cations of sera from cases of chronic nephritis which did not show any very marked changes Two cases of uremia studied by Salvesen and Linder (14) had total base within normal limits

The variations of these different factors are best shown by a study of individual cases These also serve to illustrate the great influence

TABLE 6

*Variation of total base and base combining power of the different acids in individual cases of chronic nephritis**

CASE	DATE	BLOOD NON PROTEIN NITRO GEN	BASE BOUND BY PROTEIN	BASE BOUND BY INOR GANIC P	Cl	HCO ₃	DETER- MINED ACIDS	TOTAL BASE	UN DETER- MINED ACIDS
	1925	mg per cent	mM	mM	mM	mM	mM	mM	mM
35805	January 30	170	9.5	6.2	75.6	23.0	114	138	24
	February 3	210	9.8	9.1	70.6	18.7	108	136	28
	February 5	242	7.2	9.7	67.3	16.6	101	135	34
	1924								
34802	October 9	93	11.2	3.5	91.6	21.1	127	138	11
	October 14	126	13.1	5.1	79.7	21.6	119	142	23
	October 22	159	12.2	5.4	82.7	17.4	118	145	27
33049	April 26	234	9.8	7.2	86.9	12.8	117	146	30
	April 30	272	9.9	9.0	86.9	11.8	118	148	30
	1925								
35795	January 30	240	11.6	5.2	92.2	8.7	118	146	28
	1924								
35344	December 4	215	12.6	3.8	109.6	6.0	133	150	17
	December 5	223	11.6	4.2	103.8	6.3	126	152	26
39242	November 7	85	11.5	3.0	109.6	19.9	144	162	18
	November 14	85	11.6	3.7	109.8	18.8	144	150	6
35558	December 31	288	11.8	5.8	111.8	3.4	133	164	31
	1923								
14118	December 9	198			127.1	13.1			
	1924								
29796	April 7	134	9.3	3.7	93.8	15.0	122		
	April 14	164	8.5	3.4	93.4	19.7	125		
	April 23	190	9.5	5.2	87.2	24.3	126	162	36
	April 25	194	10.1	5.8	83.5	22.3	122	151	29
29636	March 18	107	11.7	4.4	107.4	13.6	137		
	March 26	133	11.5	5.5	96.5	15.7	129		
	April 18	81	12.9	3.9	88.8	26.3	132	148	16

* All determinations were on serum obtained without exposure to air except those on cases nos 14118 and 15071 where oxalated blood was saturated at 38°C with CO₂ at 40 mm tension and the plasma then obtained

chloride variations have on the degree of acidosis. A few case studies are shown in table 6. They are discussed in the order in which they occur in the table. As in the previous tables, values are expressed in millimols of base-combining capacity. Blood non-protein nitrogen is shown in the third column. The base equivalent of the chloride, bicarbonate, phosphate and protein combined is placed in column 8 under the heading "determined acids." "Undetermined acids," in column 10 is simply the difference between the total base and the determined acids.

The first patient (no. 35808) was a young man, 24 years old, who presented a typical picture of chronic interstitial nephritis. He had been vomiting almost constantly for 3 weeks and at the time of admission was obviously extremely ill. Vomiting continued, he grew progressively worse and died after being in the hospital one week. In the 3 studies which were made the total base of the serum remained constantly low. The total determined acids were low and decreased steadily at the expense of chloride and bicarbonate. In addition to the low base there was a definite and progressive increase in undetermined acids, during the week before death. The relationship of chloride to bicarbonate is worthy of special note. The chloride was extremely low, the reduction probably exaggerated by vomiting. As a result bicarbonate was not nearly as low as it might otherwise have been. In this case we should conclude that the acidosis was caused by both a decrease in total base and an increase in the undetermined acids and phosphate but was masked to a large extent by the extremely low chloride.

The second patient (no. 34802) was a woman, 65 years old, with chronic nephritis who had been vomiting quite frequently for several weeks. The blood examinations were made during the two weeks before death. Vomiting recurred at frequent intervals throughout this period. The changes in the blood electrolytes are similar to those of the first case. The total base was low but increased somewhat. During the same period the total acids decreased. This resulted in a steady increase in the undetermined acids. Increasing acidosis was not associated with increasing phosphate. In this case the acidosis was again mitigated by the low plasma chloride.

The next case (no. 33049) was a woman, 50 years old, with severe

symptoms who had been vomiting frequently for about 2 weeks. The blood examinations were made 5 days and one day before death. She also had low total base and high undetermined acids. Between the two blood examinations, in spite of the administration of 2000 cc of physiological saline subcutaneously, total base remained practically unchanged.

The next patient (no 35795) was a young woman, 26 years old with a marked exaggeration of symptoms during the preceding 3 weeks accompanied by frequent vomiting. The findings were similar to those of the last case except for the presence of higher chloride, associated with lower bicarbonate.

In another similar case (no 35344) the symptoms were of shorter duration and the vomiting less severe. This young woman, 27 years old, had had definite symptoms for only 2 weeks and had only occasionally vomited. The reduction in total base and increase in undetermined acids were less marked than those of the previous case but very low bicarbonate was again associated with high chloride.

The next case (no 39242) presumably had marked renal disease of a chronic type with no evidence of severe renal decompensation. She had been practically free from symptoms but showed arteriosclerosis, hypertension, albuminuria and very low phenosulfonephthalein excretion. Here, though the total base and acids were normal there was again a recession of bicarbonate in favor of chloride.

Case 35558 had severe symptoms for only 3 days without vomiting. The findings are in striking contrast with the studies on the first four cases presented. Total base was normal, but phosphate elevated and undetermined acids high. Under these conditions high chloride was associated with extremely low bicarbonate. The course of the disease appeared quite different from that of the preceding cases. He grew rapidly worse, gradually passed into coma and died after being in the hospital 48 hours. It is difficult to avoid the conclusion that vomiting has been the important factor causing the variations of chloride and secondarily, variations in the degree of acidosis.

The next two cases (nos 14118 and 15071) had extremely high chloride. In both cases bicarbonate was low, the sum of chloride and bicarbonate remaining within normal limits.

The last 2 cases illustrate the changes which followed sodium bicar-

bonate treatment of acidosis In the first case (no 29697) the increasing bicarbonate was associated with a decrease of plasma chloride, the total determined acids changing very little In the third examination the total base was normal but the undetermined acids high During the 2 days between the last 2 examinations the patient grew rapidly worse and died 48 hours later During this period he received no sodium bicarbonate and was not vomiting Total base, bicarbonate and chloride all decreased and the undetermined acids remained elevated

The last case presented similar phenomena Between the first and second blood examination the patient vomited for several days Within this period there was a marked fall of plasma chloride and a slight increase of bicarbonate During the 2 weeks preceding the third examination he received 56 grams of sodium bicarbonate Chloride receded as bicarbonate increased, the reciprocal alterations of the two acids were almost equimolecular After bicarbonate the total base remained relatively low

SUMMARY

It appears that a reduction of total base and an increase of undetermined acids are the most significant factors causing acidosis in chronic interstitial and arteriosclerotic nephritis, phosphate playing a somewhat less important rôle The degree of acidosis appears to be greatly influenced by variations of plasma chloride Frequently, especially in the presence of vomiting chloride may be quite low, thus freeing base to combine with bicarbonate and resulting in only a slight reduction of CO_2 With high chloride bicarbonate may be extremely low These studies emphasize the profound changes which occur in the blood electrolytes in the terminal stages of chronic nephritis

CASE HISTORIES

Case 35805 A man 24 years old had good health until 2 months before admission to the hospital but since then blurring of vision, loss of weight, headache, weakness and very frequent attacks of vomiting Three days before entering the hospital there was a sudden onset of shortness of breath and palpitation followed by swelling of the legs For 48 hours he had voided no urine He appeared quite ill with some orthopnea and hyperpnea He was pale and his face appeared puffy There was a marked albuminuric retinitis The lungs were clear His heart was

considerably enlarged and rapid. The systolic blood pressure was 226, and diastolic 148. There was slight edema of the ankles. The temperature was continually normal. His urine showed considerable albumin, a few hyaline casts and occasional red blood cells. The first arterial blood study was made the morning after he was admitted. Vomiting ceased for almost 24 hours but soon set in again and persisted with great frequency until just before death. On the second day pericarditis developed. Water and nutrient fluids were to a large extent vomited. He died after being in the hospital one week. Further examinations of the arterial blood were made 3 days and one day before death. Following the second blood examination he was given 1300 cc of saline by hypodermoclysis.

Case 34802 A woman 65 years old had had hypertension with headache and cardiac symptoms for 3 years. Some dimness of vision had been developing and later weakness and pallor. For several weeks before admission to the hospital she had been vomiting every morning. Apparently vomiting had been vomiting had been growing more frequent. She was stuporous and appeared quite uncomfortable. Respirations were deep and rapid. She showed pallor of the skin and mucous membranes and definite albuminuric retinitis. Her heart was moderately enlarged and arteries markedly thickened. The systolic blood pressure was 210 and diastolic 100. The lungs were clear. The liver was moderately enlarged. Her face appeared puffy but there was no edema anywhere. Temperature was normal until a rise 24 hours before death. She entered the hospital October 8, grew progressively worse and died October 26, 1924. The first blood study was made on the morning after admission. Vomiting recurred at frequent intervals throughout her course in the hospital. The vomitus contained no free HCl but on various examinations sodium chloride in concentrations of 2 to 4 grams per litre. Further blood studies were made on the 7th and 15th days. During the 3 days before death she was comatose with increasing respirations. She received 3 grams of sodium chloride on each of the 2 days preceding the third blood examination.

Case 33049 A woman 50 years old had not been well for several years. About a month before admission she had an acute upper respiratory tract infection and about a week later began to vomit. Vomiting grew progressively worse and she had eaten practically nothing and had taken very little fluids for two weeks. During the two weeks before entering the hospital she had definite cardiac symptoms. Her legs had been swollen for a few days but this disappeared before admission. She was said to have passed no urine for 2 weeks. She was stuporous and had a pasty yellow pallor. The respirations were deep. Her neck veins were distended and she had râles at the bases of both lungs. The heart was slightly enlarged and arteries moderately thickened. The liver was enlarged. The systolic blood pressure was 198, diastolic 110. There was slight edema about the ankles and forearms. The urine showed considerable albumin, a few red blood cells and granular casts. She grew progressively worse and died after being in the

hospital 6 days. The blood was studied 5 days and one day before death. On the day before the second blood study she received 1000 cc of saline by hypodermoclysis.

Case 35795 A girl 27 years old had always felt below par and for 3 years had complained of headache, weakness and spells of vomiting. Three weeks before admission to the hospital weakness and vomiting increased and dyspnea and pericardial pains developed. She had been vomiting once or twice each day. She looked very old for her age and was emaciated and markedly underdeveloped. She was semi stuporous, cyanotic and orthopneic. Respirations were deep. There was considerable brownish pigmentation about the face and neck, hypertension, marked arteriosclerosis and evidences of cardiac failure. Slight edema was present about the ankles. After entering the hospital she grew progressively worse, vomiting almost continuously, and died on the 4th day after a convulsion.

Case 35344 A girl 27 years old who had always had rather poor health had an attack similar to the present illness 7 years before. Two weeks before entering the hospital she began to suffer from headache, dizziness, insomnia, urinary frequency and vomiting. These symptoms grew progressively more severe. Dyspnea and palpitation developed. Twenty-four hours before admission she had a convulsion and had been comatose since. Her respirations were regular, rapid and fairly deep. There was marked pallor and puffiness of the face. Her heart and lungs were negative. The systolic blood pressure was 215, diastolic 115. The arteries were slightly thickened. Her eye grounds showed no exudate and there was no edema. The urine showed considerable albumin and many casts but no red blood cells. She gradually grew weaker and died 28 hours after being admitted to the hospital. Blood was examined at the time of admission and again 15 hours later.

Case 39242 A woman 30 years old apparently had had marked renal involvement during a pregnancy 9 years ago. Since then she had had no symptoms but her blood pressure remained moderately elevated and a marked albuminuria continued. She entered the hospital for one week for an investigation of her condition. Phenolsulfonephthalein excretion remained below 10 per cent. Two examinations of her blood were made while she was in the hospital.

Case 35558 A man 68 years old who had had very good health all of his life began to suffer from weakness and shortness of breath about a month before admission to the hospital. There were no marked changes in his general condition until 3 days before admission when he suddenly became quite irrational. There had been no vomiting. He was obese, slightly cyanotic and breathing deeply. There was marked arteriosclerosis and hypertension and moderate enlargement of the liver. His urine showed a trace of albumin and occasional red blood cell. He gradually passed into coma and died after being in the hospital 48 hours. Blood was examined shortly after admission.

Case 14188 An Italian laborer, 49 years old, had complained of headache and stomach trouble for several years. During this time he had had occasional attacks of vomiting, urinary frequency and mild cardiac symptoms but no edema. Symptoms were somewhat exaggerated just before admission to the hospital. On examination he showed chiefly a marked brownish pallor of the skin, a slight increase in the depth of respirations and marked arteriosclerosis. His eye grounds showed no hemorrhages or exudate. The systolic blood pressure was 150, diastolic 100. There was no edema. He grew rapidly worse, muscular twitchings developed and later convulsions and coma. He died after being in the hospital 10 days.

Case 15071 A woman 53 years old had complained chiefly of dyspnea, palpitation and precordial pain on exertion for over 3 years. For almost 3 months she had had weakness, anorexia and a productive cough. Three weeks before admission to the hospital she vomited after meals for several days. This was followed by a typical attack of herpes zoster. There was no marked exaggeration of her symptoms just before entrance to the hospital. She was pale and undernourished and showed marked arteriosclerosis, hypertension and albuminuric retinitis. There was no edema. Her urine showed a low specific gravity, a trace of albumin and occasional hyaline casts. During the first month in the hospital there was definite improvement in her general condition. Then vomiting set in and she grew steadily worse and died after being in the hospital 7 weeks. The blood examination recorded here was made soon after admission.

Case 29796 A man 62 years old had had a stroke 5 years before from which he completely recovered. Seven weeks before admission he began to suffer from shortness of breath on slight exertion and a little later his legs became swollen. Edema gradually became marked, involving even his hands and face. There was no history of vomiting before admission. He was somewhat pale and slightly drowsy. He looked dehydrated although edema involved almost the entire body. There was slight hyperpnea but no orthopnea. His heart was moderately enlarged but not rapid. The lungs showed signs of fluid at the bases. The liver was moderately enlarged. The arteries were definitely thickened. The systolic blood pressure was 210 and the diastolic 110. His eye grounds showed retinal hemorrhages but no exudate. The urine showed a very heavy albumin reaction, low specific gravity and little of importance in the sediment. Phenol-sulfonephthalein excretion was less than one per cent. Blood Wassermann reaction 4+. Throughout his course in the hospital there were recurring attacks of vomiting. Four blood examinations were made. Between the first and second he received 21 grams of sodium bicarbonate, and between the second and the third 25 grams. During the 3 weeks he was in the hospital there was little change in his general condition until just before the last blood examination. Sodium bicarbonate administration and vomiting had stopped. Respiratory distress became extreme and there was evidence of a rapidly failing circulation. He died.

48 hours after the fourth blood examination Autopsy showed marked arteriosclerosis of the kidneys

Case 29636 A man 38 years old was admitted to the hospital with a history of long standing hypertension with attacks of cardiac failure His symptoms just preceding admission were chiefly cardiac He showed anemia, hypertension, cardiac enlargement and evidence of cardiac failure, without edema His eye grounds showed hemorrhages and exudate He failed to improve at first, growing progressively weaker and developing gastric symptoms with nausea and vomiting These symptoms disappeared after a few days and he began to improve The first blood examination was made the morning after admission, the second 8 days later During the 16 days preceding the third blood examination he received 56 grams of sodium bicarbonate (5 grams daily for 4 days and 3 grams daily thereafter) He was discharged definitely improved after being in the hospital one month The patient died at home about one month later

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OBSERVATIONS ON THE EXTRINSIC NERVOUS CONTROL OF THE AURICLES AND VENTRICLES IN COMPLETE AURICULO-VENTRICULAR BLOCK IN MAN

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(Received for publication August 17, 1925)

The influence of the extrinsic nerves of the heart on the sino-auricular node is well known (9a). In addition there is probably some direct effect on other areas in the heart. With an uninjured conducting system, however, there are obvious difficulties in finding out how much of the extrinsic nerve control is exerted on the auricles and how much on the ventricles. A study of the extrinsic nervous mechanism in complete auriculo-ventricular block is probably one of the best ways to attack this problem. Cullis and Tribe (2) working on the hearts of rabbits, found that after section of the auriculo-ventricular bundle, stimulation of the vagus produced no effect on the ventricles, whereas in the auricles the normal response was elicited. By the use of atropine and pilocarpine they proved that no vagus fibres pass to the ventricles, and that the vagus effects in the intact heart were merely transmitted along the ordinary conduction paths. On the other hand, they found by direct stimulation of the sympathetic and the use of epinephrin that the ventricles have a rich sympathetic supply quite independent of the auricular innervation. In dogs, the evidence seems to point in the same direction. Wiggers and Katz (9b) found, by the use of epinephrin, that the accelerators have a specific effect on the ventricular musculature, reducing the contraction period. Eyster and Meek (3) were able to show, by paralyzing the vagus before and after excision of the sino-auricular node, that the greatest chronotropic effect of the vagus is upon the sino-auricular node.

In studies on man, recourse must obviously be had entirely to

drugs The literature on the subject consists mainly of reports of single cases, and the effect of the drug used, rather than the possible significance as to innervation Thus Hardoy and Houssay (5), Strisower (8), and Feil (4) have reported that injections of epinephrin produce an increase in both the auricular and ventricular rates, in some cases the increase in ventricular rate exceeding by a considerable number of beats the increase in auricular rate The vagal control has been studied by paralyzing the vagus endings with atropin Apparently, then, the aim of previous investigations on the human subject was the study of the drug effects and their possibilities as therapeutic agents in heart block rather than the study of cardiac innervation Therefore, we determined to investigate the nerve supply to the heart in a series of cases of complete heart block, using drugs only as a means for this end

Eight cases of complete auriculo-ventricular block were selected from the wards of Bellevue Hospital In order to study the sympathetic innervation varying doses of epinephrin were given to five patients and observations including clinical signs, blood pressures, and electrocardiograms were made By giving 3 to 5 mg of atropin sulphate the vagus endings can be paralyzed and results which are then free from any vagal influence can be noted¹ In four patients such an amount of atropine was given and observations similar to those mentioned above were made We attempted to stimulate the vagus endings, in one instance, by a full body weight dose of digitals In order to see how much influence the vagus ordinarily exerted on the accelerator impulses, each of three patients was given an injection of epinephrin (which had previously produced certain effects) while the vagi were blocked by atropin

The normal auricular and ventricular rates and blood pressures of eight cases of complete heart block are shown in table 1 The figures indicate the rate and blood pressures at the times when the patients were at rest It is interesting to note that, as death ap-

¹ We are using the term "vagus" synonymously with "parasympathetic nerves" We realize that both the sympathetic nerve trunk to the heart and the vagus contain both sympathetic and parasympathetic types of nerve Atropin paralyzes the parasympathetic endings both in the vagus and in the sympathetic trunk

proached, the ventricular rate in patient I dropped from 30 to 9 per minute, while in patient IV it rose from 34 to 84 per minute. The average age of the eight patients is 62 years, the maximum being 73 years and the minimum 49 years. The average ventricular rate is 31 per minute, the highest being 84 and the lowest 9 per minute. The average auricular rate is 79, the highest being 93 and the lowest 62 per minute. The blood pressure ranged from a systolic pressure of 240 mm Hg to 124 mm Hg and a diastolic pressure of 90 mm Hg to 50 mm Hg, the average being a systolic pressure of 162 mm Hg and a diastolic pressure of 67 mm Hg.

TABLE 1
Summary of cases

CASE NUMBER	AGE	SEX	AURICULAR RATE	VENTRICULAR RATE	BLOOD PRESSURE	
					Systolic	Diastolic
					mm.Hg	mm.Hg
1	56	M	65-88	9-30	240	80-90
2	69	M	75	30	124	50
3	73	M	62	28	130	60
4	58	M	75	34-84	124	50
5	57	M	64	34	162	68
6	49	F	72	31*	145	62
7	68	F	93	36	204	80
8	70	M	83	44	228	164

* Total ventricular rate—includes four extrasystoles

EFFECT OF EPINEPHRIN

Epinephrin was administered in doses varying from 1 to 2 mg. We avoided intravenous administration because of the fleeting action, accompanied by undesirable symptoms. Previous to the injection of the drug control electrocardiograms and blood pressure readings were made. Following the injection, tracings were taken at frequent intervals.

A summary of the findings in seven cases is recorded in table 2. The data indicate that subcutaneous doses of 1 to 2 mg of epinephrin increase the auricular and ventricular rates considerably, an increase which is not always associated with a uniform rise in blood pressure. There is usually a greater change in the ventricular rate

than in the auricular On three occasions the rise in auricular rate was preceded by a moderate fall

Figure 1 shows a typical effect of the injection of epinephrin It can be noted that whereas the ventricular rate rises from 34 to 48 (a third over "normal") there is only a slight increase (68 to 70) in the auricular rate Thus, in this instance, stimulation of the sympathetic causes the greater effect on the ventricles and very little effect on the auricles This may be explained in one of two

TABLE 2
Summary of epinephrin effects

CASE NUMBER	EPINEPHRIN	NORMAL AURICULAR RATE	MAXIMUM AURICULAR RATE AFTER EPINEPHRIN	MINUTES AFTER INJECTION	NORMAL VENTRICULAR RATE	MAXIMUM VENTRICULAR RATE AFTER EPINEPHRIN	MINUTES AFTER INJECTION	CONTROL BLOOD PRESSURE		BLOOD PRESSURE, MAXIMUM		MINUTES AFTER INJECTION	REMARKS
								Systolic	Diastolic	Systolic	Diastolic		
	<i>mgm</i>												
1	5 0	62	79	20	16	51	10	—	—	—	—	—	
2	1 0	75	128	3	30	86	3	124	50	190	140	3	
5	1 0	64	86	83	34	50	44	162	68	185	60	59	
6	1 0	71	No rise	—	32	48	35	140	64	No rise	—	—	(1)
6	2 0	78	No rise	—	29	50	10	146	58	176	56	5	(2)
7	1 5	91	132	15	36	66	15	204	80	230	75	25	(3)

Remarks

- (1) Numerous ventricular premature contractions 10 min after injection which persisted for about one hour
- (2) Ventricular premature contractions more numerous Persisted for almost three hours
- (3) Many ventricular premature contractions 5 min after injection Persisted for 35 min

ways (1) The sympathetic supply to the auricles may be relatively less effective as compared to the ventricles, or (2) the vagus inhibition may be stronger in the auricles and so prevent full sympathetic action on them To check this last possibility it is necessary to paralyze the vagus fibres completely and then to give epinephrin Such a procedure was carried out and is reported under the heading, "Effect of epinephrin after paralysis of the vagi by atropine"

No significant changes in the length of the Q R S interval in the electrocardiogram were noted after the administration of epinephrin,

so it may be possible that conduction in the ventricle is unaltered after injection of epinephrin. The R-T interval, however, was distinctly shortened. Our results do not permit us at present to make a definite statement in this regard and further work is being done on this aspect of the problem.

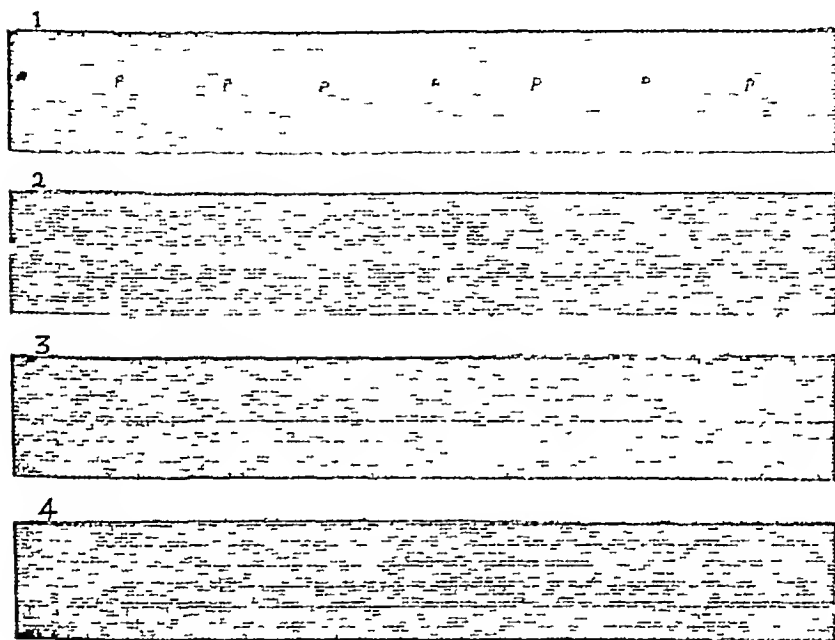


FIG. 1. EFFECT OF EPINEPHRIN. CASE 5

1, Control (normal) Lead II. 2, 4 minutes after injection of 1 mg. of epinephrin. 3, 10 minutes after injection of epinephrin. 4, 20 minutes after injection of epinephrin.

At no time did we succeed in changing complete block to a normal rhythm or to partial block.

In cases 6 and 7 there were, in the control records, occasional ventricular contractions, which did not occur in regular sequence with the dominant ventricular rhythm. Attempts were made to correlate these with auricular beats but such a correlation could never be shown to exist. We were therefore forced to assume that

these contractions were ventricular premature beats arising from a second ventricular focus. After the injection of epinephrin, the number of these aberrant ventricular complexes increased, together with an augmentation in the complexes of the dominant rhythm. The increase in number of the ventricular ectopic beats suggests that epinephrin causes an increase of irritability in the ventricle.

Exact quantitative measurements of changes in irritability and conductivity can be made only with difficulty in man, since mechanical as well as electrical events must be studied. Experimental work covering this point will be reported subsequently.

The data presented show, therefore, that the dominant effect of epinephrin is on the ventricle. Since epinephrin stimulates the sympathetic endings it seems reasonable to suppose that the sympathetic innervation of the ventricle is greater than that of the auricle.

TABLE 3
Summary of atropine effects

CASE NUMBER	ATROPINE SULPHATE mgm	NORMAL AURICULAR RATE	MAXIMUM AURICULAR RATE	MINUTES AFTER INJECTION	NORMAL VENTRICULAR RATE	MAXIMUM VENTRICULAR RATE	MINUTES AFTER INJECTION	NORMAL BLOOD PRESSURE		BLOOD PRESSURE AT HEIGHT OF ACTION		MINUTES AFTER INJECTION
								Systolic	Diastolic	Systolic	Diastolic	
2	5.0	76	100	54	30	38	34	114	56	Unchanged		65
5	5.0	70	100	15	34	38	15	190	60	140	70	
6	5.0	66	94	30	27	34	40	148	64	Unchanged		
7	3.5	98	118	10	34	36	10	205	78	Unchanged		

EFFECT OF ATROPINE

We administered 5.0 mg of atropine sulphate in four cases and 3.5 mg in another. The results of our observations are summarized in table 3. An analysis of the data indicates that, following the injection of doses of atropine sufficiently large to completely paralyze the vagus endings, the auricular rate increased by an average of twenty five beats per minute. The rise in auricular rate is practically the same as Crawford (1) found in normal individuals after paralysis of the vagi. This would indicate that the influence of the vagus on the sinus node is the same in the cases of complete heart

block studied as in normal persons. There is also a slight but distinct increase in the ventricular rate. However, we found slight variations in the ventricular rate in these patients from time to time before the drug was given. Since the increase in ventricular beats after atropine is within these control limits it is possible that we are still dealing with these slight variations even after paralysis of the vagi. Yet, the change in rate after atropine is always in the same direction, i.e., upward, and it may be that the change in rate is an atropine effect. It is possible, then, that the ventricle has a few vagus fibres which functionate even though conduction from auricle to ventricle is completely blocked. In any case, the vagus effect on the ventricle is so very slight that it can be regarded as negligible.

EFFECT OF EPINEPHRIN FOLLOWING THE PARALYSIS OF THE VAGI BY ATROPINE

We have shown above that epinephrin in small doses produces but a slight rise in the auricular rate, while the ventricular rate is markedly increased. In fact, in case 6, a dose was found which produced a moderate increase in the ventricular rate and practically no change in the auricular rate. Furthermore, from observations on the effect of atropine we were led to believe that in complete block the direct influence of the vagi on the ventricles is slight. If this is correct, one would expect that if a dose of epinephrin, such as would not produce a change in auricular rate, be administered when the vagi have been paralyzed with atropine both the auricles and the ventricles would respond with an increase, but the increase in ventricular rate would be only slightly greater than if epinephrin had been administered alone. Observations have demonstrated the correctness of this assumption in two instances (cases 6 and 7). In both cases epinephrin was administered at the time when the full effect of atropine had developed. In both patients the auricles responded with a marked rise. The increase in ventricular rate was practically the same as in the two control observations where epinephrin was administered alone. These facts are illustrated in Table 4 which shows the comparative effects of paralysis of the vagi and stimulation of the sympathetic in patient 6. When atropine alone was given, the maximum auricular rate was 94 beats per minute, thirty

minutes after injection. Thus it is evident that in table 4 (C) epinephrin was given at the height of the atropine action.

These patients were all over 40 years of age, and it might be said that their vagus tone had diminished to such an extent that blocking the fibres with atropine had no effect. The increase in auricular rate noted above when atropine was administered is evidence that such was not the case.

TABLE 4

Effect of the administration of epinephrin after paralysis of the vagi by atropine (case 6)

TIME IN MINUTES	AURICULAR RATE	VENTRICULAR RATE	PREMATURE VENTRICULAR CONTRACTIONS PER MINUTE	TOTAL VENTRICULAR RATE
A Effect of atropine				
	66	24	3	27
	Atropine sulphate 5.0 mgm injected subcutaneously			
10	80	28	0	28
17	91	29	0	29
20	91	28	0	28
30	94	28	0	28
40	93	26	8	34
50	93	27	2	29
60	88	25	5	30
80	82	26	1	27
90	82	26	0	26
100	82	27	2	29
120	80	27	0	27
B Effect of epinephrin				
	72	27	4	31
	71	26	4	30
	Epinephrin 1 mgm (1:1,000) injected subcutaneously			
2	68	25	8	33
3	68	26	8	34
4	68	28	7	35
5	72	30	2	32
10	69	28	17	45
20	67	28	15	43
30	67	28	12	40
35	72	24	24	48
45	74	28	17	45
55	71	28	8	36
65	71	27	15	42

TABLE 4—*Continued*

TIME IN MINUTES	AURICULAR RATE	VENTRICULAR RATE	PREMATURE VENTRICULAR CONTRACTIONS PER MINUTE	TOTAL VENTRICULAR RATE
C Effect of atropine and epinephrin				
	75	25	1	26
	78	26	1	27
	76	26	1	27
	Atropine sulphate 5 mgm injected subcutaneously			
5	74	25	2	27
10	90	29	0	29
15	94	29	0	29
20	95	28	0	28
25	93	26	0	26
30	95	27	0	27
35	Epinephrin 1 mgm (1 1,000) injected subcutaneously			
37	108	30	2	32
40	118	32	9	41
45	123	31	12	43
50	119	32	12	44
55	115	30	14	44
60	106	28	12	40
70	108	29	8	37
80	108	29	8	37
90	105	27	9	36
110	103	26	10	36

The evidence, then, indicates that the experimental results of Cullis and Tribe are in the main applicable to man

EFFECTS OF DIGITALIS

In other forms of cardiac irregularities, digitalis has been used to stimulate the vagus endings, particularly in reference to the auricles. With this in mind, a full body-weight dose of a standardized tincture of digitalis was given to patient 6. Frequent clinical observations were made and many electrocardiograms taken during the next twenty-four hours. No change in the auricular or ventricular rates occurred, although the patient was completely digitalized as evidenced by the slight nausea of which she complained and the inversion of the T wave in Lead II of the electrocardiogram.

EFFECT OF EXERCISE

Many observations have been made in the past showing that in complete block there is no rise in the ventricular rate after exercise. In order to make this study as complete as possible, we observed the effects of exercise in our patients. Two instances are reported below.

Patient 6 exercised by bending the knees and body twenty-five times. As shown in table 5, there is an immediate rise in the auricular rate, about the same as one would expect in a normal individual,

TABLE 5
Effect of exercise (case 6)

TIME	AURICULAR RATE	VENTRICULAR RATE	PREMATURE VENTRICULAR CONTRACTION	TOTAL VENTRICULAR RATE	BLOOD PRESSURE	
11 35	78	26	2	28	130	62
11 40	76	25	2	27	134	60
11 46-47 Exercise—25 Knee-bends						
11 48	104	23	7	30		
11 52	82	20	10	30	145	64
11 55	81	19	11	30	135	62
12 00	85	27	7	34		
12 05	81	28	4	32		
12 13-14 Exercise—25 Knee-bends						
12 15	108	21	7	28	150	62
12 20	94	27	5	32		
12 25	92	27	8	35	130	64
12 30	96	29	5	34		

but no change in the ventricular rate. Patient 4, on his first admission, when at rest, showed an occasional dropped ventricular beat without an increased P-R interval as his only evidence of interference in conductivity between auricles and ventricles. Upon moderate exercise (bending the body backward and forward about twenty times) his rhythm, as shown by the electrocardiograms, changed to a complete heart block with long periods of asystole. Such a change from a partial to a complete block on exercise has also been noted by Sands (7).

EFFECT OF CHEYNE-STOKES RESPIRATION

The frequent association of Cheyne-Stokes breathing with complete heart block has been noted by a number of observers who apparently observed no abnormal changes in heart rate with different phases of respiration. In 1913 Oppenheimer and Williams (6) reported a case of complete a-v block with Cheyne-Stokes respiration in which, during the dyspneic period, the auricular rate was strikingly reduced as compared with the period of apnea. The ventricular rate, however, was only slightly lowered during dyspnea. The explanation offered was that the vagus in complete heart block retained a marked chronotropic effect on the auricle and little, if any, on the ventricle. This explanation is in accord with the conclusions which we drew from the epinephrin and atropine experiments. However, in our series, patient 3 showed a marked slowing of ventricular rate at the beginning of the hyperpneic period and an increase in ventricular rate during the apneic period. The auricular rate was unchanged throughout. In the light of all previous work this could not be a vagal effect. It is possible that, in this instance, the anoxemia resulting from the apnea affects the ventricular muscle directly. In two other patients with Cheyne-Stokes breathing no auricular or ventricular rate changes were noted which could be correlated with respiratory phases.

At least three conditions may therefore occur during Cheyne-Stokes breathing in complete block, as shown by these and other cases reported in the literature:

1. No change in the auricular or ventricular rates.
2. A marked slowing of the auricular rate during the dyspneic period and a corresponding rise during apnea, but very little change in the ventricular rate.
3. A marked slowing of the ventricular rate during the dyspneic periods and a corresponding rise during apnea, but no change in the auricular rate.

VARIATIONS IN AURICULAR AND VENTRICULAR COMPLEXES

In the course of this work our attention was directed to the frequency with which variations in auricular and ventricular complexes

of the electrocardiograms were encountered. The drug employed, the dosage, and the time of injection seemed in most instances to bear no relation to the occurrence of these abnormalities, in fact they were frequently present in the control tracings. Possibly the same factors which produced the complete block are operative here. Some

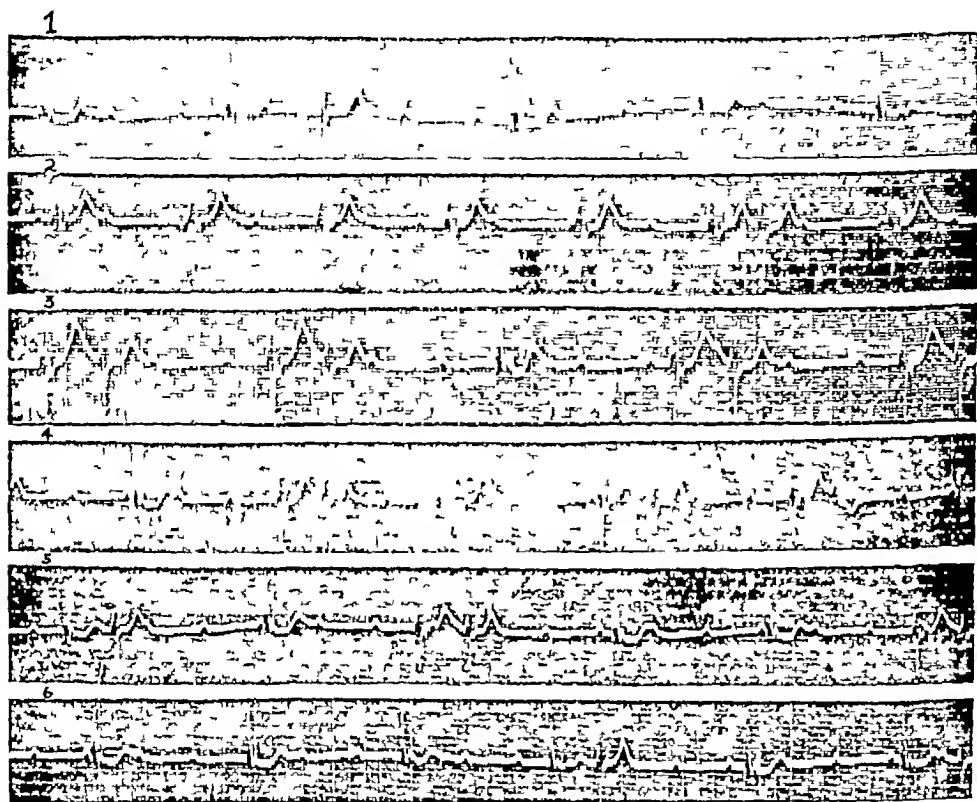


FIG 2 EFFECT OF EPINEPHRIN UPON THE VENTRICULAR COMPLEXES CASE 6

1, control electrocardiogram 2, electrocardiogram at height of epinephrine effect 3, 4, 5, 6, successive steps in return to normal

of the abnormalities of the P wave noted were inversion, dropping and irregular spacing

Two cases showed intraventricular block in addition to their a-v block. Two cases presented ventricular premature contractions of reversed electrical potential from the dominant ventricular potential. No regularity in the occurrence of these beats was noted although ventricular ~~arrhythmia~~ was paid to it, several five minute tracings having

been taken and carefully counted. The ventricular premature contractions in these cases apparently came from a focus fairly high in the conducting system because the R bases throughout were less than 0.10 second.

In case 6, the ventricular premature complexes were in the opposite direction to the normal dominant ventricular complexes. Fifteen minutes after the injection of epinephrin (on two occasions) the direction of all the complexes was that of the premature contractions (tracing 3, fig. 2). Accompanying these changes there was considerable precordial pain. The ventricular complexes returned to the original type 165 minutes after the injection of epinephrin.

Three possible explanations may be presented.

1 That epinephrin during the height of its activity changed the electrical axis of the heart.

2 That, with the increase in irritability of the ventricular musculature after the administration of epinephrin, the focus which had previously been responsible for only an occasional ventricular premature systole, now became the dominant pacemaker of the ventricle, and that as the effect of the epinephrin wore off the original pacemaker gradually assumed control again.

3 That epinephrin produced a change in conduction. None of these theories satisfactorily explain tracing 3. Although in tracing two, all the complexes have the same direction, yet the rhythm is not regular and complexes are present which appear to be premature in relation to other complexes just as was the case before epinephrin was given. If we are to conceive of all the ventricular complexes in tracing two as coming from one focus, then we would have to assume a varying irritability. It is even more difficult to think of a change in the electrical axis where the reversion back to normal is accomplished by a gradual increase of normally directed complexes as the effect of the drug wears off. If the explanation were an alteration of ventricular conductivity as a whole one might expect variations in the duration of the QRS group. These are not apparent.

SUMMARY

An effort was made to study the extrinsic nervous control of the auricles and ventricles in the human heart. Eight cases of complete

heart block were studied with epinephrin, atropine, and digitalis. Observations were also made as to the effect of Cheyne-Stokes respiration and exercise.

Subcutaneous injections of epinephrin in doses varying from 1 to 5 mg were given in seven cases. A considerable rise in ventricular rate always occurred and usually a rise in auricular rate, which, however, was not as great. The blood pressure usually rose but in one case the rise was transient and was followed by a fall. In two instances the rise in ventricular rate was associated with the appearance of numerous ventricular premature contractions probably from some focus other than the one originating the dominant rhythm. Since it is generally assumed that the effect of epinephrin on the heart is similar to a stimulation of the sympathetic endings, we have indications from these observations that the sympathetic control of the ventricles is more pronounced than that of the auricles.

Atropine sulphate in doses sufficient to paralyze the vagus endings was administered in four cases. The auricular rate increased by an average of 25 beats per minute, an increase which agrees with the rise of 23 beats per minute found by Crawford in normal individuals after paralysis of the vagi. This would indicate that the influence of the vagus on the sinus node is the same in the cases of complete block studied as in normal persons. A slight but distinct rise in ventricular rate occurred in each instance, indicating that the vagi exert a slight inhibiting effect on the ventricle in complete block.

When epinephrin was injected after the vagi had been paralyzed with atropine, although the auricular rate rose considerably, the ventricular rate increased only very little. It is evident, therefore, that the vagal influence on the ventricles is slight while the sympathetic control is considerable.

Digitalis in one patient caused no variations in auricular or ventricular rates although there was full digitalization as evidenced by the slight nausea and the inverted T wave in the lead of the electrocardiogram.

The cases in which the effect of exercise was noted presented the usual findings, a rise in auricular rate, but no change in the ventricular rate.

Three patients had Cheyne-Stokes respiration. In two of them

no variations were noted in auricular or ventricular rates, while the other showed a marked slowing of the ventricular rate at the beginning of the hyperpneic period and an increase during the apneic period with no change in the auricular rate

Many variations in ventricular and auricular complexes were noted and special attention was paid to a curious reversal of the ventricular spikes after administration of epinephrin in one case

CONCLUSIONS

1 Evidence is presented to show that in complete heart block the accelerator nerves exert a considerable control over the ventricles whereas the vagi have only a slight influence

2 Vagal control over the auricles is the same in complete heart block as in the normal heart

3 Exercise in complete heart block tends to increase the auricular rate but leaves the ventricular rate unchanged

4 Cheyne-Stokes breathing in complete heart block may be associated with varying auricular and ventricular responses

5 A case is reported where the injection of epinephrin caused a complete change in direction of the ventricular complexes

REPORT OF CASES

Case 1 J L, a laborer, aged 56, was admitted to the Fourth Division medical service on August 23, 1923, complaining of frequent dizzy spells and increasing weakness. He was entirely well up to six days before admission when he began to have two to three fainting attacks per day, associated with loss of consciousness and marked weakness. The frequency of these attacks increased so that at the time of admission he had about twenty a day. He was found to be dyspneic, orthopneic, cyanotic, with dilated jugular veins but no oedema. The examination of the chest was negative. The heart showed a moderate enlargement, a systolic blow at the apex, auricular sounds in the diastolic period, and a ventricular rate of 38 per minute. It was found that whenever the ventricular rate dropped to about 12 per minute, the patient would have a period of unconsciousness and convulsions. At one time a ventricular rate of 8 per minute with a ventricular standstill of 16 seconds was noted. Epinephrin was given to this patient as a therapeutic procedure because it was found to materially lessen the convulsions. The patient died on September 12, 1923, in a "status epilepticus."

Case 2 G S, a watchman, aged 69, was admitted to the Second Division medical service on January 23, 1923, complaining of shortness of breath. He stated that he had had "heart trouble" for over fifteen years but was able to do his ordinary work until three years ago when he began to have sudden attacks of shortness of breath bearing no constant relationship to exertion. At the time of admission to the hospital he was unable to go about, the slightest exertion causing dyspnea. There were occasional attacks of dizziness but no loss of consciousness. He was found to have no dyspnea when at rest, no signs of congestive heart failure. His chest was moderately emphysematous. Examination of the heart showed a moderate enlargement, distant sounds, no murmurs, and a ventricular rate of 30 per minute. It was noted that the jugular pulse showed faint wavelets asynchronous with the ventricular beats. On February 6, 1923, the patient had a period of semi-consciousness with a few tremors of the extremities. The heart rate at that time was 10 per minute. After that several periods of unconsciousness with convulsions occurred daily. He was discharged on March 29, 1923. On October 2, 1923, he was again admitted following an attack of dizziness and unconsciousness, during which he had fallen. His physical condition was practically the same as on the previous admission. He was again discharged November 2, 1923.

Case 3 A L, a barber, aged 73, was admitted to the Second Division medical service on December 11, 1923, in marked failure with Cheyne-Stokes breathing. It was impossible to obtain any history other than that his illness had begun about one year before with shortness of breath. He was very dyspneic, orthopneic, with marked cyanosis of all extremities. There was no oedema, although, the jugular veins were engorged and filled from below. The heart was moderately enlarged, the sounds were not very clear, but a loud high pitched systolic murmur was heard over the entire precordium. The urine showed a marked amount of albumen. He became steadily worse and died on December 15, 1923.

Case 4 W M, a floor polisher, aged 58, was admitted to the Third Medical Division on July 7, 1923, complaining of shortness of breath and dizzy spells followed by short periods of unconsciousness. These symptoms began about February, 1923. He remained in a hospital for four weeks and was discharged improved. Just before this admission his attacks of syncope began to recur every 20 to 30 minutes. He was neither dyspneic or orthopneic. There were no signs of congestive failure. There was marked sclerosis of all the peripheral vessels. The heart was moderately enlarged, with sounds of fair quality at the apex. The rhythm was regular at times, at other times there were dropped beats every third or fourth cycle. After exercise the condition became one of complete block with long periods of asystole. He was discharged improved on August 2, 1923. In December 9, 1923, he was readmitted in coma with Cheyne-Stokes breathing and a heart rate of 60 to 85 per minute. An electrocardiogram showed complete heart block even with this rapid rate. He died on December 20, 1923.

An autopsy showed a calcified band across the intraventricular septum, along the base of the mitral valve

Case 5 J L, a watchman, aged 57, was admitted to the Third Medical Division on November 16, 1923, after having been found unconscious in his room a few minutes before. He regained consciousness soon after he was brought to the ward. He stated that this was his first attack, and that previous to this he had been in the best of health. He was found to be dyspneic on slight exertion, but had no congestive failure. All the peripheral arteries were markedly sclerosed. His heart was only slightly enlarged. There was a systolic murmur at the apex and also at the aortic cartilage. The rhythm was regular, the rate being 46 per minute. He was discharged on December 1, 1923, somewhat improved.

Case 6 C D, a housewife, aged 49, was admitted to the Third Medical Division on December 1, 1923. Her history dates back to August 1922, when she had a period of unconsciousness lasting about three minutes following unusual and severe exercise. About a month later she began to have frequent mild attacks of dizziness. In February 1923, she fell unconscious on the street without any warning, and had considerable difficulty in getting home. The next day she had a loss of consciousness with convulsions and was taken to a hospital where she remained for several weeks having frequent convulsions together with marked slowing of the pulse. Since then she has practically lived in hospitals being unable to do any work whatsoever. At rest, she presented no signs of failure. The heart was moderately enlarged, with a rough systolic murmur at the apex, sounds of good quality and a rate of 26 per minute. There were a few premature contractions. She was discharged on February 9, 1924 with very little, if any improvement.

Case 7 E T, an embroider, aged 68, was admitted to the Third Medical Division on January 5, 1924, complaining of fainting spells, dyspnea, and palpitation. She was very unstable mentally and a reliable history could not be obtained. She had had ten to fifteen fainting spells since October 1923 and had been in bed most of the time since then. Her heart showed a slight enlargement, and a ventricular rate of 38 per minute while the jugulars pulsated at a rate of 84 per minute. The vessels were moderately sclerosed. She was discharged unimproved, January 27 1924.

Case 8 L M, a cobbler, aged 70, was admitted to the Second Medical Division on February 26, 1924, in coma with convulsions, after having been picked up in the street. He had a large scalp wound over the mid occipital region. There was no bleeding from the nose, ears or mouth. Neurological examination was negative. The heart was slightly enlarged to the left. The rate was 24 per minute and regular, the sounds were rather blurred, there were no murmurs.

The pulse was of the Corrigan type. He regained consciousness for only a few days and then lapsed into a state of coma with Cheyne-Stokes breathing, which continued almost to the day of his death, March 13, 1924.

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THE EFFECT OF CHANGES IN REGIME UPON THE URINARY ALKALINE TIDES IN A NORMAL INDIVIDUAL

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(Received for publication December 10, 1925)

In 1912 Henderson and Palmer (1912) introduced a technique for determining the pH of the urine and applied it to the study of twenty-four-hour specimens from 222 normal and pathological cases. Their observation showed a range of pH from 5.0 to 7.4. They also observed that the pH of the urine rises to some extent following a meal. The pH curve of normal urine was also studied by Hasselbalch (1912) who reported a fall in pH immediately after each meal followed by a rise to a maximum about three hours later. Fiske (1921) confirmed the existence of this rise in pH after meals and observed pH values during the alkaline tide as high as 7.7. He studied also the phosphate excretion during the period of the alkaline tide. Hubbard, Munford and Allen (1924) observed that the alkaline tide occurred only in individuals who showed presence of free HCl in the stomach during digestion. Higgins (1914) and Erdt (1915) found a rise of alveolar $p\text{CO}_2$ during digestive activity. Van Slyke, Stillman and Cullen (1917) confirmed this observation and found that the plasma bicarbonate in some cases increases slightly, in others not. Dodds (1921) and Dodds and McIntosh (1923) observed a rise in the CO_2 content of the blood and in alveolar $p\text{CO}_2$ about one-half hour after meals followed by a secondary fall in both at the time the urinary alkaline tide occurs.

Leathes (1919) found that normal active individuals given water without food at 8 a.m. showed an increased urinary alkalinity during the morning hours. He attributed the alkalinity to increased pul-

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monary ventilation incident to their activity. He observed also that in cases of acute and subacute nephritis this morning alkaline tide was diminished or absent. McCorvie (1925) has studied the morning alkaline tide following the ingestion of water by the method of Leathes and also the urinary pH and phosphate excretion in 8 normal and 74 pathological cases. He confirms the observation of Leathes that the morning alkaline tide characteristic of normal cases tends to disappear in nephritic cases.

The following study is presented because it furnishes data upon the pH curve of a healthy subject throughout the entire day under varying conditions as regards time of taking food and time of commencing the day's activities. First the curve of urinary pH is shown when in accordance with the individual's custom only two meals, morning and evening, were taken. Then the curve when three meals were taken and then with only a single meal at midday. Finally the curve is shown during a day when the subject took his usual breakfast, but remained quiet in bed until noon without any physical activity.

METHODS

The urinary pH was measured by the nitrophenol method of Michaelis. The preparation of the standards in this method depends upon the fact that the indicators are colored when alkaline and colorless when acid. The standards consist of a series of tubes containing alkaline aqueous solutions of the indicators in concentrations selected to give the same depth of color as that of a series of buffer solutions containing a uniform concentration of dye.

Two indicators are used: Paranitrophenol (pH 5.4 to 7.0) and metanitrophenol (pH 6.8 to 8.4).

The solutions employed are as follows:

- | | |
|-------|--|
| No. 1 | 0.1 per cent aqueous solution of paranitrophenol |
| No. 2 | 0.3 per cent aqueous solution of metanitrophenol |
| No. 3 | $\frac{1}{10}$ M/10 aqueous solution of NaHCO_3 |
| No. 4 | 5 cc. of no. 1 diluted to 50 cc. with no. 3 |
| No. 5 | 5 cc. of no. 2 diluted to 50 cc. with no. 3 |

The standard tubes are made as shown in table 1.

A pH determination is made as follows: To 2 cc. of urine is added

4 cc of 0.9 per cent sodium chloride solution and 1 cc of indicator solution 1 or 2. Comparison with the standard is made in a Walpole colorimeter block, a tube containing 2 cc of urine plus 5 cc of saline being placed behind the standard tube.

The same series of standards permits one to measure also the pH range from 5.2 to 5.0. For this purpose one takes 2 cc of urine, 2.5 cc of salt solution, and 2.5 cc of indicator solution 1. A color matching the standard tube marked pH = 5.4 will then represent a pH of 5.0 and one matching the tube marked pH = 5.6 will represent a pH of 5.2.

TABLE 1

pH	SOLUTION 4	SOLUTION 3	pH	SOLUTION 5	SOLUTION 3
	"	"		"	"
5.4	0.16	6.84	6.8	0.27	6.74
5.6	0.25	6.75	7.0	0.43	6.57
5.8	0.40	6.60	7.2	0.66	6.34
6.0	0.63	6.37	7.4	1.00	6.00
6.2	0.94	6.06	7.6	1.50	5.50
6.4	1.40	5.60	7.8	2.30	4.70
6.6	2.08	4.92	8.0	3.00	4.00
6.8	3.00	4.00	8.2	4.20	2.80
7.0	4.05	2.95	8.4	5.20	1.80

The color of the standard tubes was checked by comparison with Sorensen's phosphate buffer solutions to which the nitrophenol dyes were added. The pH determinations were made within a few hours of obtaining the urine except that the evening specimens at 8 and at 10 o'clock were kept on ice until the next morning. The subject's urine kept even 24 hours on ice was never observed to show a change of pH greater than 0.1.

RESULTS

The subject was a healthy male aged 28 years. His urine was collected at two-hour intervals from 8 a.m. to 10 p.m. and its pH determined. His daily work was light in character in the hospital and laboratory and was constant from day to day.

The first period of study was of the urinary pH curve of the subject during twenty consecutive days during which as was the custom of the

subject only two meals morning and evening were taken In the first four days the diet was as follows

Breakfast at 8 30 a m A cup of coffee, two fried eggs, two slices of bread and butter

Supper at 6 p m Vegetable soup, Hamburger steak, coffee

During the next fifteen days it was approximately the same

TABLE 2
Urinary pH at two-hour intervals for twenty days Two meals daily

	8 A M	10 A M	12 N	2 P M	4 P M	6 P M	8 P M	10 P M
1st day	6 1	6 3	6 8	6 5	5 7	5 6	5 9	6 4
2nd day	6 0	6 0	7 0	6 5	6 5	5 8	5 9	5 9
3d day	5 7	5 9	6 6	6 0	5 6	5 8	5 9	6 2
4th day	5 7	5 9	6 4	6 3	5 4	5 3	6 0	6 3
5th day	5 8	5 6	6 4	6 2	5 6	5 6	6 3	6 0
6th day	6 1	5 8	6 6	5 4	5 4	5 5	6 2	6 3
7th day	5 9	5 9	6 6	5 9	5 6	5 9	6 2	6 0
8th day	5 6	5 4	6 4	5 6	5 3	5 8	6 3	6 2
9th day	5 5	5 4	6 3	5 8	5 3	5 2	6 0	6 0
10th day	5 2	5 3	6 2	5 2	5 2	5 6	6 0	5 8
11th day	5 6	6 1	6 4	5 2	5 4	5 4	5 8	5 8
12th day	5 4	6 2	6 3	5 4	5 5	5 9	6 0	5 8
13th day	5 8	5 4	6 3	5 4	5 2	5 4	6 0	5 8
14th day	5 6	5 4	6 6	6 0	5 8	5 8	6 3	5 9
15th day	5 6	5 4	6 2	5 8	5 3	5 4	5 9	5 6
16th day	5 8	5 8	6 3	5 8	5 4	5 4	5 8	5 8
17th day	5 9	5 7	6 3	6 1	5 5	5 3	6 0	6 2
18th day	5 5	5 7	6 5	5 6	5 2	5 3	5 9	5 7
19th day	5 6	5 4	6 3	6 0	5 8	5 8	6 0	6 2
20th day	5 7	5 7	6 3	5 9	5 4	5 3	6 0	6 1
Average	5 7	5 7	6 4	5 8	5 5	5 5	6 0	6 0
Minimum	5 2	5 3	6 2	5 2	5 2	5 2	5 8	5 6
Maximum	6 0	6 3	7 0	6 5	6 5	5 9	6 3	6 4

Breakfast, 8 30 a m—Supper, 6 00 p m

In table 2, and in figure 1 are shown the curves of the average, the maximum, and the minimum, of the pH at each time of day for twenty days Each day's observations showed alkaline tides after the meals, the morning tide having its maximum about $3\frac{1}{2}$ hours after breakfast the evening tide about 2 hours after supper The morning alka-

line tide was followed by a period of maximal acidity. The curve also shows that the alkaline tide after breakfast was higher than that following supper.

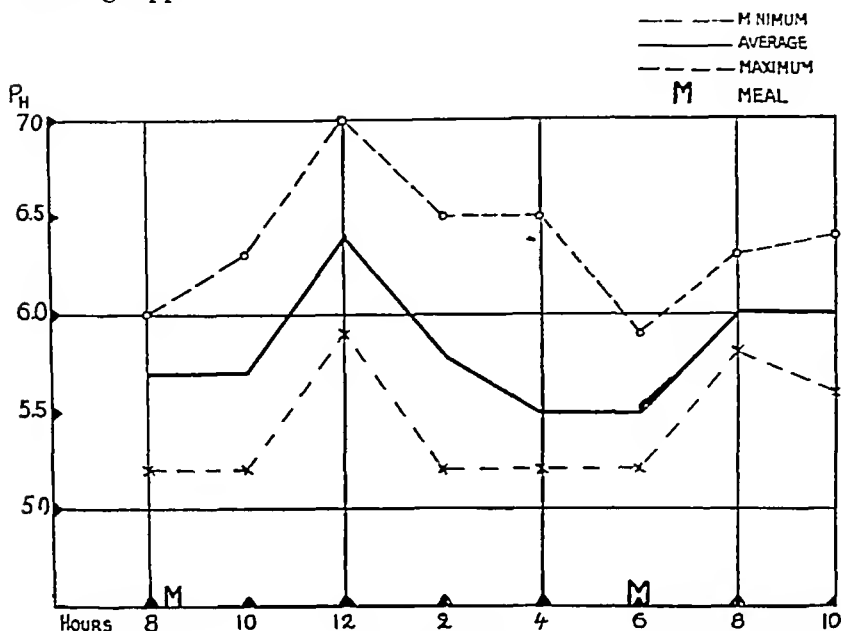


FIG 1 URINARY REACTION CURVES THROUGHOUT THE DAY SHOWING TWO ALKALINE TIDES

Based on observation over twenty consecutive days (The minimum at 12 is drawn to include the low figure from table 3, May 1)

TABLE 3
Urinary pH at two-hour intervals on three days Three meals daily

	8 A.M.	10 A.M.	12 N.	2 P.M.	4 P.M.	6 P.M.	8 P.M.	10 P.M.
April 29, 1925	6.0	5.8	6.0	6.6	6.6	5.6	6.8	6.5
May 1, 1925	5.8	5.5	5.9	5.4	5.5	6.2	6.2	5.9
May 5, 1925	5.5	5.9	6.8	6.4	5.4	5.6	5.7	6.1

Breakfast at 8.30 a.m., Lunch at 1.00 p.m. Supper at 6.00 p.m.

The second period of study was of the pH curve when the subject took three meals daily. The results on these days are shown in table 3 and figure 2. On each of these days there are only two alkaline

tides, never three and on only one of the three days is an alkaline tide seen after the noon meal when it occurs apparently as a delay of the tide usually occurring in the morning. The evening rise tends perhaps to be higher than during the two-meals-per-diem period.

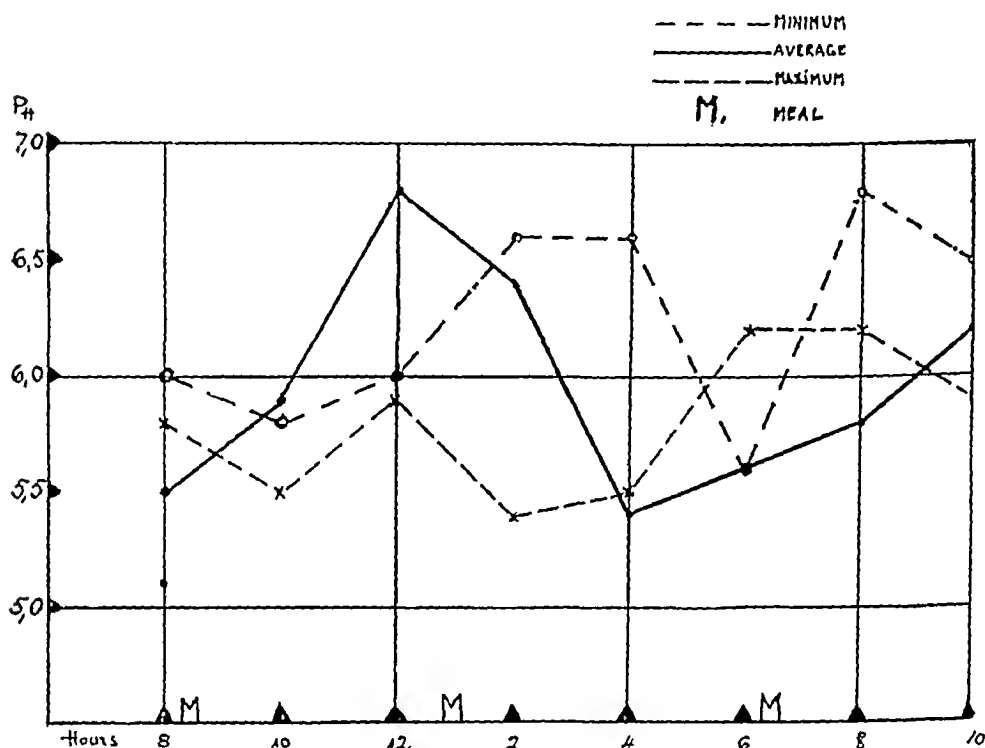


FIG 2 SHOWING URINARY pH WITH THREE MEALS DAILY, WITH TWO ALKALINE TIDES

Very similar to those in figure 1

TABLE 4
Urinary pH at two-hour intervals on three days One meal daily

	8 A.M.	10 A.M.	12 N.	2 P.M.	4 P.M.	6 P.M.	8 P.M.	10 P.M.
May 6, 1925	5.9	7.5	8.0	7.0	6.4	5.8	6.1	5.4
May 12, 1925	5.9	6.3	7.2	7.0	7.0	6.0	6.8	6.0
May 20, 1925	5.8	5.5	6.4	6.2	5.4	5.2	5.4	5.9

Meal at 1 00 p.m.

The third period of study was of the pH curve when the subject took only one meal per day at 1 p.m. The results on these days are

shown in table 4 and figure 3. Again two tides are in evidence. In two instances the morning tides without breakfast are very pronounced. In only one instance is there a suggestion of prolongation of a high pH following the one o'clock meal.

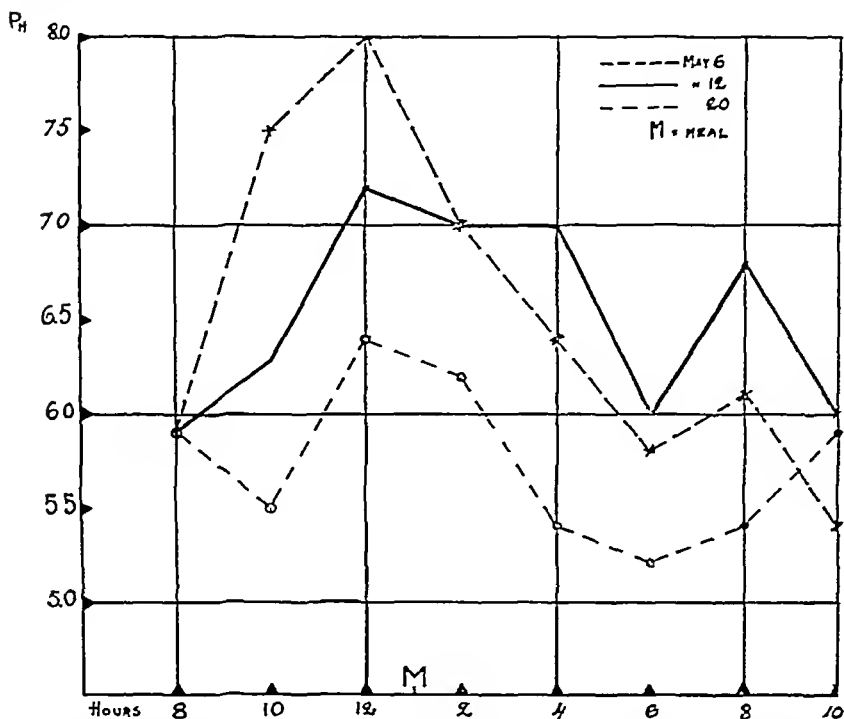


FIG 3 SHOWING URINARY pH WITH ONE MEAL DAILY WITH TWO ALKALINE TIDES

Very similar to those in figure 1

The fourth study was to determine the effect of physical activity on the alkaline tide by keeping the subject in bed until noon. The result is shown in figure 4. Two tides are seen, timed approximately the same as upon other normal or experimental days.

These studies suggest that activity and the period of digestion while they may perhaps influence the urinary pH are not the sole cause of the rhythmic alkaline tides in the urine. They strongly

suggest a rhythm in the urinary pH characteristic for the individual and tending to persist in spite of quite striking alterations in his regime

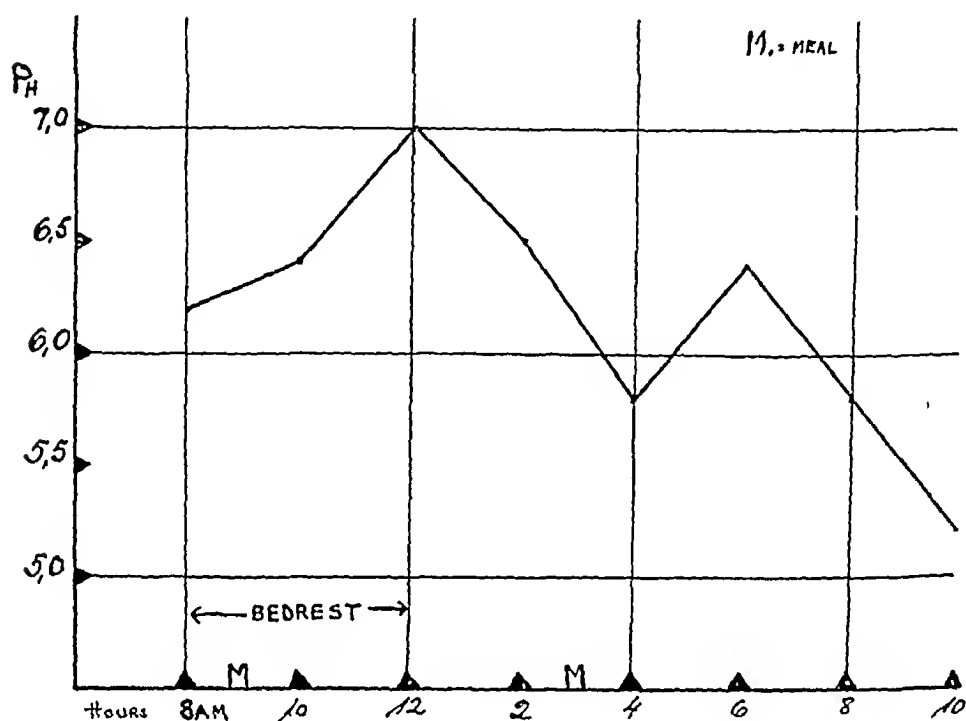


FIG 4 SHOWING URINARY pH WITH REST IN BED UNTIL 12 NOON WITH TWO ALKALINE TIDES, UNAFFECTED BY ABSENCE OF ACTIVITY

SUMMARY

Determinations of the pH of the urine of a healthy subject were made at two-hour intervals throughout the day on twenty-seven days

For twenty days the subject according to his habitual custom took only two meals, morning and evening

For three days he took three meals daily

For three days he took only one meal daily at 1 p m

On one day he remained in bed until noon, taking his usual breakfast but with no physical activity

The curves of urinary pH on all of the days showed two alkaline tides, one with its maximum about noon and the second with its maximum about 8 p m

The intensity and timing of these alkaline tides was not conspicuously altered by the changes in the patient's regime

The observations suggest a rhythm in urinary acidity characteristic for the individual and persisting in spite of marked variations in the individual's regime

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THE EXCRETION OF AMMONIA AND TITRATABLE ACID IN NEPHRITIS

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(Received for publication December 28, 1925)

As the result of past metabolic studies, especially those of Henderson and Palmer (12) it is known that the non-volatile acids produced in excess of fixed base by human metabolism are excreted in the urine in two forms, viz, as free acids, and as ammonium salts

Since the kidney is unable to form urine with a pH much lower than 5.0, it can excrete, in significant amounts, free acids of only the weak buffer type. In this class fall acid phosphates and the various organic acids. It appears, however, that free acid excretion may assist also in elimination of strong acids, which can react with buffer salts (e.g., $\text{HCl} + \text{Na}_2\text{HPO}_4 = \text{H}(\text{NaHPO}_4) + \text{NaCl}$), the free buffer acid being excreted in place of the strong acid. Thus Marriott and Howland (19) found that HCl ingestion increased the output of free buffer acids.

Ammonia serves to neutralize either weak or strong acids. Consequently both ingestion of HCl, (either as such, or as NH_4Cl or CaCl_2) and diabetic ketosis (producing weak beta-hydroxybutyric acid) cause increase in ammonia excretion. In fact both the above conditions increase both ammonia and free buffer acid output, without greatly altering the NH_3/acid ratio (Marriott and Howland (19), Fitz and Van Slyke (4)). Peculiarly, however, ingestion of acid phosphate was found by Marriott and Howland to increase only the output of titratable acid, without any effect whatever on the ammonia output.

Since the nature of the acid eliminated, and presumably other unknown factors, can influence the proportions excreted as ammonium salts and titratable free buffer acids respectively, it is not surprising

that, as shown by Henderson and Palmer, and by our data, wide fluctuations occur in the NH_3/acid ratio even in the same normal individual. To take an extreme case, when a vegetarian diet produces an alkaline urine the NH_3/acid ratio may approach infinity, because the acid factor in the denominator approaches zero, while the ammonia in the numerator, although greatly reduced, is still measurable.

Nevertheless the ammonia in a series of urines from an individual with a normal excretory mechanism ordinarily averages from 1 to 2.5 times the free acid, and this ratio is maintained over great ranges of total $\text{NH}_3 + \text{acid}$ output, e.g., from a low normal 300 cc. of 0.1 N $\text{NH}_3 + \text{acid}$ per 24 hours to the 4,000 or 5,000 cc. that may be excreted in diabetic acidosis. There is apparently some tendency towards a moderate increase of the ratio in the latter condition. (In the data of Fitz and Van Slyke (4) the normal urines show, as do the normal data of the present paper, a mean NH_3/acid ratio of approximately 1.5 while the mean for the cases with ketosis is 2.2. This difference in the ratio is small, however, considering the immense range of total $\text{NH}_3 + \text{acid}$ values covered.) The eliminated acid is divided between ammonium salts and free acid with a sufficient degree of constancy to justify the definition of the usual limits of the NH_3/acid ratio in the urine of normal individuals, and the assumption of an abnormality in the acid excreting mechanism when this ratio consistently lies outside such limits. A persistent abnormally high ratio may result from bacterial formation of ammonia from urea in the bladder. A persistent abnormally low ratio is presumably a sign of damage to the ammonia-forming function, particularly when the titratable acid factor of the ratio appears unaffected.

Henderson and Palmer (12, 11) determined the free acid by titrating the urine to pH 7.4, the reaction of normal blood plasma. The NH_3/acid ratio¹ averaged over a number of days in the urine of normal subjects varied from 0.7 to 2.0. Nephritic cases fell into 2 groups.

¹ Henderson and Palmer expressed their results by the ratio acid/ NH_3 . We have inverted this ratio and use the expression NH_3/acid , because (1) the relative rise and fall in the ammonia appears to be the significant factor in varying nephritic conditions, the free acid excretion being relatively unaffected. With the ammonia in the numerator, therefore, the ratio rises and falls with the more

Among the cases with normal ratios there was a predominance of conditions diagnosed as degenerative nephritis. In another group, more sharply definite in regard to urinary character, the NH_3/acid ratio varied from 0.3 to 0.6, these low values being due to diminished ammonia output. On the average in this group of cases the urine volume was abnormally great, its pH unusually low, and the "total acid output" ($\text{NH}_3 + \text{acid}$) diminished because of deficit in the NH_3 constituent. The low urinary pH was interpreted to indicate a tendency towards nephritic acidosis, presumably caused by the reduced ammonia formation. In this group chronic glomerulonephritis was prominent.

Stillman, Van Slyke, Cullen, and Fitz (31) reported from this hospital a case of acute nephritis following pneumonia in which there was almost complete suppression of ammonia excretion, and a progressive fall in blood alkali reserve. When the latter had dropped to about half normal, the clinical condition began to improve and the ammonia output gradually increased until it reached the unusual maximum of 1500 cc. of 0.1 N ammonia per day. The daily ammonia excretion then also fell to the usual 300 or 400 cc., presumably because there was no further need for unusual acid elimination. (Only the alkali reserve data were published, the present ammonia figures are from the hospital records.)

Rabinowitch (25) has recently shown that in diabetic patients with albuminuria the NH_3/acid ratio averages lower than in normal persons or in diabetic patients without albuminuria. He concludes that diabetics with injured kidneys have a subnormal ability to form ammonia, and are probably rendered thereby unusually susceptible to acid intoxication.

Interest both in the origin of the ammonia of the urine and in its variation in nephritis has been augmented by Nash and Benedict (21) who have shown that the ammonia is probably formed by the kidneys. It had been believed that the ammonia was formed else-

significant variable. (2) The NH_3/acid ratio, as will be shown, is low when kidney function is diminished, and in its relation to renal function it appears simpler to denote ammonia-acid relationship by an expression which parallels the renal function rather than by one which increases when the function falls.

where in the body and carried by the blood to the kidneys. The early determinations of ammonia in blood, with the exception of those of Folin and Denis (5), were compatible with this hypothesis. Nash and Benedict found that when minute precautions were taken the blood from the carotid and vena cava of dogs contained less than 0.1 mg of ammonia nitrogen per 100 cc. The blood from the renal vein, however, contained about twice as much, a fact which strongly suggested that the kidneys are the site of the ammonia formation. Loeb Atchley and Benedict (18) have repeated and confirmed these observations.²

In this paper we present the results of routine determinations of the NH_3 /acid ratio carried out on nephritic patients over a period of several years. (Many of the patients reported in this paper have been included in previous papers on other phases of nephritic metabolism (14, 15, 16, 17). Blood ammonia determinations on a number of patients and on normal subjects have been made to determine whether the low ammonia excretions observed in the former were due to diminished formation or to retention.

An attempt has been made to correlate the variations in the NH_3 /acid ratio with the type and severity of the cases. The latter have been classified according to the plan of Volhard and Fahr (15, 35, 36) in four groups, nephrosis, acute glomerulonephritis (stage I), chronic glomerulonephritis (stages II, and III), and nephrosclerosis. Each type is reported in a table by itself, and in each table the cases are arranged as far as possible in the order of increasing severity, as judged by the clinical state and renal function tests. We realize that the Volhard-Fahr types are not separated by sharp lines, and that opinions are likely to differ in classifying certain cases. We have given in the tables such data as space permits to assist the reader in forming his own judgment.

² Strauss (32), using the vacuum distillation method of Hahn and Kootz (9) reported the normal blood ammonia nitrogen as 0.8 to 2.0 mg per 100 cc and nephritic values ranging up to 6 mg. Gherardini (8) reported 0.4 to 0.7 mg in both normal and nephritic subjects. Such high values seem explainable only by errors of technique.

EXPERIMENTAL

The analyses of nephritic urine were carried out on 24-hour specimens which had been preserved with toluene in an ice box from the time the individual specimens were obtained. In some cases 24-hour urine specimens were examined daily. In others they were taken every second day or once each week. In the series of determinations on normal individuals the specimens were collected over periods generally of about 2 hours, and analysed immediately.

Methods Urinary ammonia was determined as described by Van Slyke and Cullen (33), 5 cc of urine being mixed with 5 cc of saturated potassium carbonate solution and aerated into standard acid.

The *titratable acidity* of the urine was determined by Folin's familiar method (6). Because the neutrality of our solid potassium oxalate could not be trusted, we have used instead of the solid substance prescribed in the original method, a saturated solution neutralized to phenolphthalein. Fifteen cubic centimeters of this solution and 0.5 cc of 1 per cent phenolphthalein solution were added to 25 cc. of urine, the mixture was thoroughly shaken, and was titrated to a pink color (pH approximately 8.5) with 0.1 N NaOH. Henderson and Palmer (12, 11) used neutral red as indicator and pH 7.4 as end point, a procedure which has an advantage in physiological interpretation in that the results express the amount of alkali required to bring the urine to the pH of normal blood serum. For the purpose of ascertaining the NH_3/acid excretion ratio, however, we have preferred the phenolphthalein titration because its more alkaline end point represents a urinary reaction at which the ammonia excretion becomes very small. Even in urine alkaline to phenolphthalein there is, it is true, some ammonia, and in consequence a urine with a pH of 8.5 would yield an NH_3/acid ratio of infinity. When the amount of 0.1 N alkali required for 25 cc of urine falls below 1 cc. the NH_3/acid ratio may accordingly be higher than in urine of more usual acid content excreted by the same individual. The interpretation of the physiological and clinical significance of the ratio in such urines is therefore uncertain. They are, however, rare.

No attempt has been made to prevent escape of CO_2 from the urine. We have preferred to perform the titrations under conditions that

would be a practicable part of hospital routine, and to permit such increase in the margin of variation as might result. In any but the most alkaline urines the effect could not be important.

The blood ammonia Five cubic centimeters of oxalated blood were transferred, within three minutes of the time of drawing, into the test tube of the Van Slyke-Cullen apparatus used for urea estimations, in which the rubber connections were as short as possible. An equal volume of saturated potassium carbonate, from which the ammonia had been almost completely removed by aeration for one hour, was added. A flowmeter (22) was connected, and the contents of the tubes were aerated for 5 minutes with an air current flowing at the rate of 5 liters per minute. (Van Slyke and Cullen found that with this time and air current 98 per cent of the ammonia was removed.) The ammonia was caught in a tube, calibrated at 25 cc, containing 1 cc of 0.005 N acid diluted to about 15 cc with ammonia-free water. As indicator we used 4 drops of a saturated solution of methyl red in 70 per cent alcohol, as suggested by Barnett (2). At the end of the aeration the tubes were washed down and diluted to 25 cc with ammonia-free water. The excess acid was titrated with 0.005 N alkali to a standard end point color, by which all the solutions had been standardized. For this end point a buffer mixture of pH 5.6, made by mixing 91 cc $\frac{M}{5}$ sodium acetate and 9 cc $\frac{M}{5}$ acetic acid, was used. The standard color was that of 25 cc of this mixture to which 4 drops of indicator had been added. The titrations were made from a 2 cc micro burette, so that the volumes delivered could be measured to within 0.005 cc. Blank determinations were performed on the reagents, and the amount of ammonia obtained from them was deducted from that found in the blood analyses. The accuracy of this method is shown in table 1.

Table 2 shows that all of the measurable ammonia of the blood is aerated in the first 5 minutes when the rate of air flow is 5 liters per minute (controlled by a flowmeter). Further aeration, up to 40 minutes, gave no more ammonia, which proves that the potassium carbonate did not react with any constituent of the blood to produce ammonia. In fact the carbonate prevents the spontaneous ammonia formation that occurs in drawn blood. This spontaneous formation,

TABLE 1

Recovery of ammonia from standard ammonium sulfate solutions by the blood ammonia method

NH ₄ -N present per 100 cc. of solution	Amount of solution used for analysis	NH ₄ -N found per 100 cc.	Mean value for NH ₄ -N found per 100 cc.	Recovery
mg	cc	mg	mg	per cent
0 171	5	0 173 0 160 0 160 0 173 0 187 0 173	0 171	100
0 085	5	0 076 0 070 0 070 0 063	0 07	82 4
0 060		0 05 0 05	0 05	83 4

TABLE 2

Effect of aeration time and delay in analysis on blood ammonia determination

Sample number	Aeration time	Blood NH ₄ -N per 100 cc.	Average blood NH ₄ -N per 100 cc.	Remarks
	min	mg	mg	
1	First 5	0 014 0 020	0 017	Determinations made 5 minutes after blood drawn
	Additional 5	0 0	0	
	Additional 10	0 0	0	
	Additional 20	0 0	0	
	5	0 328 0 342	0 335	Same blood 1½ hours after drawn
2	5	0 021 0 014	0 017	Determination made 5 minutes after blood drawn
	5	0 042	0 042	Determination made 15 minutes after blood drawn

already noted by Parnas and Heller (23, 24), makes it important to add the carbonate to the blood immediately on drawing. The ammonia value may be doubled when the blood stands 15 minutes without addition of carbonate. If the blood is analyzed after standing 1½ hours the increase may be 17 fold.

Blood urea determinations were made according to the method of Van Slyke and Cullen (33).

The urea concentration index was calculated from the urea content of blood and urine by a modification of the original method of Austin, Stillman, and Van Slyke. The present index³ is calculated as

$$\frac{\text{Urine urea concentration}}{\text{Blood urea concentration}} \times \sqrt{\text{cc urine volume output per minute}} = \frac{U}{B} \sqrt{V}$$

The simple concentration ratio $\frac{U}{B}$ represents the number of times the kidney concentrates the urea in compressing it from blood concentration, B , to urine concentration, U , when the urine volume output is the average normal 1 cc per minute (= 1440 cc per 24 hours). When the urine volume is less than 1 cc per minute, the concentration U will normally be greater, and vice versa. In order to correct for this volume effect we multiply the simple ratio $\frac{U}{B}$ by the square root of the volume, an empirical correction which was found to hold for ordinary urine volumes (under 2 cc per minute) by Austin, Stillman, and Van Slyke. In order to apply the formula to subjects of different weights, we have used the *volume per kilo body weight*, $\frac{V}{W}$,

³ The present formula is identical with that of Austin, Stillman, and Van Slyke, but is rearranged into concentration terms for convenience in calculation and interpretation. The former equation was $K = \frac{D}{B\sqrt{VW}}$, where D is the output of urea per time unit, B , V , and W having the same significance as above. The output, D , is the product of the volume, V , and the concentration, U . Hence $D = UV$. Substituting UV for D we have $K = \frac{UV}{B\sqrt{VW}} = \frac{UV}{B\sqrt{VW}} = \frac{U}{B} \sqrt{\frac{V}{W}}$. A more detailed account of the use of the concentration index will be given shortly in another paper (20).

instead of the absolute volume. In this case we have used the cc hourly output as V , since for adults of average size, W is about 60, and $\frac{\text{cc per hour}}{\text{body weight}}$ approximates cc per minute.

The concentration index $\frac{U}{B} \sqrt{V}$, or with the weight correction $\frac{U}{B} \sqrt{\frac{V}{W}}$, varies from 35 to 80 in normal subjects. Occasionally a single determination as low as 30 may be encountered, but we have observed no normal subjects with an index repeatedly and consistently below 35. That is, the normal adult, excreting urine at the average normal rate of 1 cc per minute, concentrates urea in the urine to at least 35 times the blood concentration, usually between 40 and 60. The numerical values of the index with the present units of volume are 6.5 times as great as the values with the units used in the Austin Stillman, Van Slyke index, which varied in normal subjects from 4.5 to 10.5.

The *phenolsulphonephthalein* output was determined as described by Rowntree and Geraghty (27). The injections were made intravenously, and a fresh standard was prepared for each test.

The serum CO_2 content was determined according to the method of Van Slyke and Neill (34).

The serum pH was determined according to the method of Hastings and Sendroy (10).

To avoid reporting individually a large number of observations on patients, the daily acid and ammonia excretions have been averaged for periods of several weeks or months. Changes in diet, treatment, or in the state of renal function were taken as reasons for starting new periods.

ANALYSIS OF RESULTS

Normal subjects (Table 3 and fig. 1). Ninety-five determinations of the ammonia-acid ratios were made on 11 normal subjects. The collection of specimens was generally made over 2-hour periods in the course of the day. No attempt was made to control the diet, fluid intake, or activity of these subjects. The volume, specific gravity, and the absolute amounts of ammonia and acid excretion varied consider-

TABLE 3—Continued

Number	Subject	Date	Time	Volume	NH ₃ excreted per hour	Acid excreted per hour	$\frac{\text{NH}_3}{A}$
				cc	cc 0.1 N	cc 0.1 N	ratio
3	J C B	4/ 8/25	2 30- 4 00	55	25 9	14 9	1 74
			4/ 9/25	2 00- 3 33	42	29 9	16 8
		4/16/25	11 45- 1 45	64	24 7	14 1	1 75
		4/17/25	10 30-12 00	116	18 0	7 3	2 47
			12 00- 2 10	118	20 2	12 5	1 62
		4/23/25	6 40- 9 00	143	23 0	12 5	1 84
			9 00-11 00	157	12 4	3 7	3 35
			11 00- 1 00	78	15 2	11 8	1 29
			1 00- 3 00	52	21 5	14 7	1 46
			3 00- 4 30	43	14 3	9 1	1 52
		4/24/25	7 15- 9 15	60	19 0	10 7	1 78
			9 15-11 15	73	19 6	6 7	2 93
			11 15- 3 15	165	37 5	18 3	2 05
Average							1 97
4	J W	4/11/25	9 00-11 00	107	34 7	13 8	2 52
		4/16/25	8 00-10 00	435	139 0	144 0	0 96
			2 00- 4 30	72	30 7	26 6	1 15
		4/17/25	1 00- 2 30	111	34 0	35 1	0 97
Average							1 40
5	F C	4/11/25	9 00-11 00	64	22 2	7 5	2 96
		4/16/25	10 30-11 05	33	26 8	13 7	1 96
			3 00- 4 00	40	26 3	12 6	2 09
		4/17/25	9 20-11 20	78	17 7	7 5	2 36
Average							2 34
6	W N	4/6/25	11 00- 1 00	360	29 9	18 6	1 61
			1 00- 3 00	290	42 7	33 6	1 27
			3 00- 5 00	265	38 4	24 2	1 59

TABLE 3—Continued[illegible]

TABLE 3—*Concluded*

Number	Subject	Date	Time	Volume	NH ₃ excreted per hour	Acid excreted per hour	$\frac{\text{NH}_3}{\text{A}}$
9	J A P	4/ 9/25	2 05- 4 05	79	cc 0.1 N 23 2	cc 0.1 N 22 4	ratio 1 03
			4/11/25	9 25-11 30	110	19 4	12 7
		4/16/25	10 10-11 45	76	14 9	10 9	1 37
			11 45- 2 00	100	21 8	20 6	1 06
		4/17/25	6 50- 9 03	132	14 5	4 2	3 45
			9 03-11 25	137	11 2	4 1	2 73
		4/23/25	6 55- 9 15	124	32 2	13 3	2 42
			9 15-11 15	105	15 5	6 8	2 28
			11 15- 1 25	77	17 7	21 5	0 82
			1 25- 3 30	63	25 6	30 7	0 83
		4/24/25	6 50- 9 20	130	21 0	14 0	1 50
			9 20-11 20	86	13 6	5 6	2 43
			11 20- 1 35	70	15 8	18 7	0 85
			1 35- 3 35	65	25 1	23 2	1 08
Average							1 67
10	T N	4/16/25	11 30-12 30	185	73 4	53 8	1 36
			1 30- 3 30	69	19 6	19 7	1 00
		4/17/25	8 30-10 30	374	106 2	132 6	0 80
			10 30- 3 00	184	16 1	14 5	1 11
Average							1 07
11	S S	4/16/25	1 15- 3 15	82	12 3	16 3	0 75
		6/3/25	10 10-11 45	67	10 6	6 5	1 63
			11 45- 1 45	140	8 3	6 8	1 22
Average							1 20

ably The ratios varied between the extremes 0.6 and 4.9, but the majority fell between 0.8 and 2.5. The mean individual ratios fell between 0.9 and 2.3, the majority lying between 1.0 and 2.0, similar to those of Henderson and Palmer (13). Normal individuals show for ammonia and acid excretion the wide flexibility which seems char-

acteristic for normal kidney function. For this reason, isolated estimations are of little value, the variability of the function is important as well as the value of the ratio

Nephrosis (Six cases, table 4 and fig 1) The patients in this

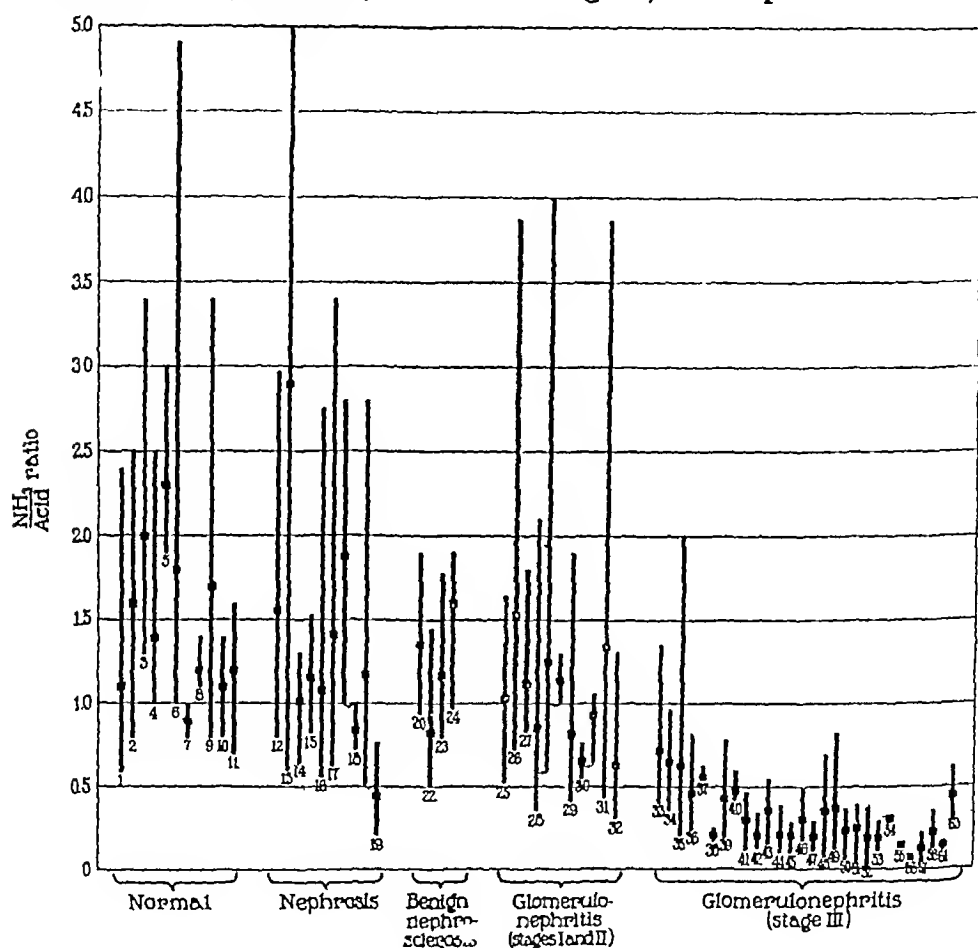


FIG 1 The lines represent the entire range of ammonia-acid ratios for each case. The solid blocks represent the mean ratios. Two or more lines joined by a bracket represent different states of renal function in the same patient, as explained in the text. The lines are numbered to correspond with the case numbers in the tables.

group showed normal values for the ammonia-acid ratio. There was the usual variation in range for the individual and the averages. The ratios varied from 0.56 to 5.0, the majority falling between 0.6 and 2.8. The average of the ratios for the individual cases varied from 1.02 to 2.9, all but one lying between 1.0 and 2.0.

In B S, 13, the ammonia was always high and the acid excretion moderate, consequently, the ratio was high. During the period from March 25 to April 18 large doses of calcium chloride were given after the method of Blum, Aubel and Hausknecht (3), there was a rise in ammonia output which reached a maximum of 1000 cc. 0.1 N solution. This ammonia formation presumably was the protective response of the organism to prevent acidosis from the HCl absorbed from the administered CaCl_2 (Salvesen, Hastings and McIntosh (29)). The calcium chloride had no effect on the edema. Between May 20 and June 14 a high protein, low fat diet was given. The protein was increased from 40 to 80 grams, and thence by successive additions to 140 grams, i.e., 4.4 grams per kilo body weight. Analysis of the diets of Sherman's tables (30) shows that the excess of acid radicles over basic was increased from 80 cc. of decinormal acid to approximately 400 cc. An increase in the output of acid and ammonia followed, but, whereas the calcium chloride produced a large increase of ammonia and a small increase of acid, the high protein diet produced a smaller output of ammonia but a larger increase of acid. A year later, during a relapse, both the titratable acidity and the ratio were still high.

In another case, M R (17), the administration of 5 to 10 grams of calcium chloride daily had a similar effect in increasing the ammonia-acid ratio, due to the excess excretion of ammonia over acid radicles. In this patient the pH and bicarbonate of the serum remained normal as a result of this compensatory mechanism.

In another case, G H, 14, the lack of effect of sodium chloride is shown, where the ratio remained fairly constant, as did the absolute figures.

In nephrosis, therefore, the kidney function as regards ammonia and acid excretion is normal. This finding is in accord with the normal ability of the kidney to excrete and concentrate urea.

Table 5 shows 2 cases, B F, and S J, which came under observation with some of the characteristics of nephrosis, and later apparently developed renal insufficiency. In B F, probably complicated by some other underlying condition, possibly Addison's disease, a change during the course of observation was indicated by a fall in the index of urea concentration and phthalein output. A definite drop in the ammonia-acid ratio, with a tendency to fixation, ran parallel with these findings, although the ratio was still within the lower limit of the normal. The case S J was under observation during two different admissions to the hospital. During the first period the ammonia-acid ratio was over 1, during the second period there were signs of definite functional impairment, and the ratio had fallen to 0.54, and finally to 0.32, at which time there was a fixation at that low value. The change in the variability of the ratio during these two periods is strikingly illustrated in figure 1, where the first column for this patient shows a fluctuation of ratio from 0.52 to 2.8, and the second from 0.24 to 0.76.

AMMONIA AND ACID EXCRETION

TABLE 4
Cases of nephrosis

Number	Case	Age yrs	Body weight edema free kg	Period of observation	Blood pressure	Blood urea N per liter gm	Urea index $\frac{U}{V} \sqrt{\frac{B}{V}}$	Phthalein in 2 hours per cent	Plasma CO ₂ content mM	Plasma pH	Protein intake per day gm	NH ₃ excretion per day cc 0.1 N 0.1 N	Acid excretion per day cc 0.1 N 0.1 N	$\frac{NH_3}{A}$ ratio	Remarks
12	F R	20	56	1921 November 5-December 11		0 08	58 8	60			60-90	354	162	2 20	Acute nephrosis
				1921-1922 December 21-April 23	98/65	0 05	44 0 64				40-70	197	180	1 10	
				1922 May 5-May 23		0 06	36 8				60-70	342	139	2 46	
				1922 November 5-December 27	95/70	0 07	77 3 69				60-70	313	124	2 53	
13	B S	11	33	1923 March 25-April 8		0 09	102 2				40	691	171	4 04	Recovered CaCl ₂ therapy
				May 20-June 14		0 06	82 6				80-140	451	206	2 19	
				1924 February 17-March 16	108/75	0 23*	70 4 70	22 4 7	35		45-50	266	50	5 32	
				March 18-April 16		0 12*	130 0				60-70	301	116	2 60	
14	G G	24	55	1925 January 4-January 10	126/62	0 09	90 7 67	7			50	307	219	1 40	Recovered Urea 30 gm daily Urea 20-10 gm daily
				1925 January 29-February 26	130/78	0 145	37 8 62	4 31	27 38		60	263	268	0 98	
				February 27-March 11		0 124	35 0 65	3			75	199	178	1 12	
				March 12-March 21		0 101	34 6 68	1			75	218	243	0 90	
				March 27-April 16		0 102	35 1 60	6			90	233	219	1 06	Chronic nephrosis NaCl free diet NaCl, 10 gm daily NaCl free diet

15	W J	27	65	1925 March 13-May 2	120/72	0 05	85 2/76 8				65	264	231	1 14	Urea 30-60 gm daily
16	B B	23	56	1924 February 13-March 10 March 11-May 28		0 08 0 31*	56 8/70 59 3	31 9/7 34			50 80	157 256	110 252	1 12 1 02	
17	M R	29	55	1924 December 1-December 28 December 29-February 12 February 13-March 3 March 12-April 6 April 16-May 19	106/68 106/66 110/74 116/78 120/78	0 09 0 16 0 17 0 07 0 09	39 4/75 43 5/72 40 8/60 28 2/65 31 9/60	31 6/7 46 30 5/7 41 31 3/7 37	60 100 125 60 75	142 219 194 134 645	79 384 480 116 307	1 80 0 57 1 03 1 16 2 10	CaCl ₂ 5-10 gm daily		

* During urea therapy

TABLE 5
Cases first observed with clinical signs of nephrosis, and later developing renal insufficiency

Number	Case	Age	Body weight edema free	Period of observation	Blood pressure	Blood urea N per liter	Urea index $\frac{U}{V} \sqrt{\frac{B}{IV}}$	Phthalein in 2 hours	Serum CO ₂ content	pH	Protein intake per day	NH ₃ excretion per day	Acid excretion per day	$\frac{NH_3}{A}$	Blood NH ₃ -N per 100 cc	Remarks
18	B F	24	47	1925 January 27-April 14 April 14-May 19	98/70	0 10	31 5	52 31	5 7	47 35-50	312	166	1 88	0 02	Persistent edema	
					96/70	0 21	18 4	41		50-65	170	206	0 83			
19	S J	33	58	1924 January 16-February 10 February 16-June 19 December 16-January 15	120/80	0 04	39 8	59 32	0 7	38	40	150	120	1 25	Urea 30-60 gm per day	
					115/75	0 32	55 1					174	163	1 06		
					128/80	0 17	17 6	68 29	4 7	45 40-60	101	189	0 54			
				1925 January 16-February 15 February 16-March 13	128/80	0 20	16 4	69 27	2 7	49	60	128	316	0 41		
					122/70	0 18	20 9	51			60	57	176	0 32		

TABLE 6
Cases of benign nephrosclerosis

Number	Case	Age years	Body weight edema free kg	Period of observation	Blood pressure	Blood urea N per liter gm	Urea index $\frac{U}{V} \sqrt{\frac{B}{V}}$	Phthalein in 2 hours per cent	Serum CO ₂ content mM	pH	Protein intake per day gm	Ammonia excretion per day cc N/100 ml	Acid excretion per day cc N/100 ml	$\frac{VH}{A}$ ratio	Remarks
20	II Lr	29	55	1923 November 19-December 21	170/120	0.15	30.4	61	28.5	7.34	45-60	282	207	1.36	Benign
21	C T	34	74	1922 November	175/105	0.15	21.3				50	141	175	0.81	Benign
22	L J	18	54	1923 February 8-March 5	210/145	0.18	19.4	51	27.1		30-50	113	112	0.80	
23	M S	26	19	December 11-December 20 December 27-January 24 1922	196/135	0.10	25.9	37	24.1	7.36	55	237	183	1.30	
24	I R	13	70	April 5-June 8 1922	210/135	0.21	23.1	13	29.2		100	152	278	1.63	During stay in hospital apparently benign type Died July 16 in uremia after sudden onset of severe renal insufficiency

Benign nephrosclerosis (Five cases, table 6 and fig 1) All of these patients had only moderately diminished renal function as indicated by the urea concentration index. The NH_3/acid ratios varied from 0.5 to 1.9, with the averages from 0.83 to 1.6. These average values are within the limits of normality, but it is seen from figure 1 that the range of variation is much less than that of normal subjects.

Glomerulonephritis, stage I (acute) and stage II (Eight cases, table 7 and fig 1) In this group are included some patients who recovered completely, some with residual albuminuria and slight hematuria, and others with edema and urinary findings such as to warrant the possibility of transition into the chronic stages. However, in all these patients the history is definitely that of an acute onset, and the findings at the end of the period of observation are compatible with normal kidney function. The ratios in this group range from 0.31 to 4.0, the majority lying between 0.6 and 2.0, with averages from 0.62 to 1.53.

The first 2 cases, W. K., 25, and D. L., 26, were comparatively mild, yet their ratios increased during the course of recovery.

D. G., 28, was a severe case with marked edema (20 kilos over weight), low urea concentration index and phthalein output, increased blood urea, and some anemia. During this period the ammonia-acid ratio averaged 0.87, which is within the normal limits and, apparently, a discrepancy in view of the other findings. However, the ratio varied considerably, at times falling far below the normal limit. This variation is interesting in view of the ultimate outcome, because the patient suddenly eliminated all the edema, at which time the blood urea, urea index, and phthalein output, and hemoglobin returned to normal, while the ammonia-acid ratio increased to the usual normal value, without recurrence of the low figures. Further observations are desirable to determine the prognostic value of the variable but lowered ratio.

B. B., 30, was a moderately severe case which had a low normal function at the time of admission but later became normal. During the first period the ammonia-acid ratio averaged 0.65 and was decidedly fixed between 0.54 and 0.76. Later it rose to 0.93.

The output of acid and ammonia was a little low in the patient L. S., 31, during the acute stage of his illness. At the time that the edema was eliminated, the ammonia output increased and there was a temporary decrease in acidity with a rise in the ratio.

E. A., 32, showed a ratio that was persistently at the lower limit of normal, although all other functional tests were normal. Further observation is neces-

sary in order to determine the significance of such values in the face of otherwise normal functional and good clinical condition. E A may exemplify a type of case in which a fixed low normal ammonia-acid ratio is the only evidence of functional disturbance in latent glomerulonephritis following apparent recovery from acute nephritis. The urea index and phthalein output were normal.

Glomerulonephritis, stage III (chronic) and malignant nephrosclerosis. (Thirty cases, tables 8 and 9 and fig 1) This group includes patients with chronic diminution of urea concentrating power of all degrees of severity, from the early moderate forms of renal insufficiency to terminal cases with uremia and ability to concentrate only 5-fold or less. An attempt has been made to arrange them in table 9 according to the state of renal compensation and clinical condition. It is obvious that no sharp lines can be drawn on the basis of any functional tests, yet, on the whole, the milder forms of chronic nephritis have distinctly higher ammonia-acid ratios and urea indices than the more severe and uremic types. As seen from table 8 and figure 1, the first 5 cases showed moderate lowering of the urea index, phthalein output, and ammonia-acid ratio, with moderate nitrogen retention. The average ammonia-acid ratios in the first 6 patients ranged from 0.38 to 0.88, with one higher terminal value of 1.18 in J D, who died of septicemia. The remaining cases were of increasing severity. In these the ammonia-acid ratio decreased to a value as low as 0.07 in one case, M K, 56, and all were below 0.47, with the exception of S L, 49, in whom the ratio was 0.6 during one period. In the severe uremic cases, 58, 59, 60, 61, the ratio paradoxically rose to infinity in the terminal analyses due to an alkaline urine, which in turn may have been due to a terminal bacterial contamination of the urine. The patients were in coma, and the resulting urinary retention, such as occurred in 61, may have led to bacterial decomposition.

In the group of chronic glomerulonephritis with diminished urea excreting function, there was a striking fixation of the ammonia-acid ratio as well as of the urea index, the only exceptions being in the terminal alkaline urines mentioned above. It can be seen from the table that the low ratios are due to lowered ammonia excretion, never to unusually high daily acid output.

Cases V S and N H, 61 and 62, are glomerulonephritis on a sclerotic basis, which would be classified by Volhard and Fahr as malignant nephrosclerosis.

TABLE 7
Cases of glomerulonephritis, Stage I (acute) and Stage II

Number	Case	Age years	Body weight edema free kg	Period of observation	Blood pressure	Blood urea N per liter gm	Urea index $\frac{U}{V} \sqrt{\frac{1}{V}}$	Phthalein in 2 hours per cent	Serum CO ₂ content mM	pH	Protein intake per day gm	Ammonia excretion per day cc 0.1 N	Acid excretion per day cc 0.1 N	$\frac{NH_4}{A}$ ratio	Blood NH ₄ -N per 100 cc. mg	Remarks
25	W K	11	43 6	1922 February 9-March 14 March 22-April 24 May 1-June 13	100/65 95/65	0 14 0 08 0 08	54 7 45 7 94 7	43 83 59	22 9 25 4	26-40 70 70	146 175 268	195 0 183 0 206 1	75 96 30			
26	D L	14	28 32	1924 January 3-January 24 February 4-March 20 March 23-May 4	125/70 125/65	0 10 0 06 0 06	55 0 47 8 40 7	61 74			30 40 40	135 172 188	150 0 93 1 134 1	90 85 40		Initial blood pressure 165 mm
27	C C	13	38	1925 April 17-May 17	106/70	0 10	68 2	65			50	210	188 1	12		
28	D G	30	55	1924-1925 December 18-February 17 February 18-April 7 April 8-May 12	140/90 110/75 112/76	0 33 0 09 0 09	17 7 73 8 67 6	37 64 60	25 2 27 34	75 100 75	246 232 175	295 0 181 1 153 1	87 28 0 14		01	Marked edema Edema elimination Slight return of edema (nephrotic type)

Number	Case	Age	Body weight edema free	Period of observation	Blood pressure	Blood urea N per liter	Urea Index $\frac{U}{B}$	Phos. est. in 2 hours
		years	kg			mg		per cent
33	J D	14	38	1922 May 23-July 27	140/78	0 21	27 8	51
		15	41	1923 March 4-March 25	120/68	0 15	24 6	
			42	November 27-November 29	130/65	0 23	23 9	50
34	F M	13	33	1924 January 31-March 4	130-130 130-130	0 15	18 8	26
				March 6-March 30				
				April 1-May 8		0 20	10 0	31
35	A Sd	40	54	1922 March 5-March 29		0 26	17 5	49
				May 15-June 14		0 57	9 0	24
36	J O'M	16	41	1923 October 12-December 24	115/70	0 43	16 2	51
				December 25-March 27		0 19	20 7	46
37	H M	7	18	1922 April 8-April 19		0 40	18 4	15
38	M G	12	33	1925 May 27-May 30	128/74	0 12	31 0	62
39	H L	10	29 5	1922 May 10-June 5	160/110	0 18	31 6	41
			31	November 5-November 30	120/90	0 44	8 4	30
40	F P	47	61.2	1924 October 22-November 26	198/104	0 32	13 2	24
41	R N	37	58 4	1924-1925 December 12-January 17	150/100	0 21	20 0	46
				January 18-January 31	124/80	0 32*	24 4	60
				February 13-February 24	126/82	0 27	14 9	43
				March 4-March 16	115/80	0 19	17 6	47
				March 17-April 16	130/80	0 16	18 6	31
42	I C	44	51	1922 March 20-April 11	130/80	0 24	17 4	50
				May 18-June 4		0 31	16 2	49
43	P L	28	57	1925 January 13-January 29	164/90	0 31	14 8	55
				January 30-March 18	124/74	0 26	21 7	59

*During urea therapy

and malignant nephrosclerosis

pH	Protein intake per day	Ammonia excretion per day	Acid excretion per day	$\frac{\text{NH}_4}{\text{acid}}$	Blood $\text{NH}_4\text{-N}$ per 100 cc.	Remarks
	gm	cc 0.1 N	cc 0.1 N	ratio	mg	
7 29	30-50	117	157	0 75		
	70	103	162	0 64		
	10	328	277	1 18		Soda bicarbonate, 27 gm. Died December 2, 1923, of septicemia
7 34	30-35	108	154	0 70		Edema elimination
7 38	35-40	90	157	0 57		
	40	130	181	0 72		Died November 1924, of pneumonia
	40-50	83	197	0 43		
	60-70	170	193	0 88		
	50	144	250	0 58		
	60-70	114	297	0 38		
	30-50	55	98	0 56		
	35	63	321	0 20		
	50	91	209	0 43		
	45	51	169	0 30		Ascites and hydrothorax
7 40	60	112	237	0 47		Died December 6 1924
7 44	65	115	459	0 25		
7 40	65	94	430	0 22		Nephrotic type, urea, 30 gm. per day
	100	101	359	0 28		
	60	91	208	0 44		
	75	69	249	0 28	0 01	
	60	66	297	0 22		
	70	49	226	0 22		Died May 24, 1924
7 39	60	94	345	0 27		
	75	84	246	0 34	0 09	

AMMONIA AND ACID EXCRETION

TABLE I

Number	Case	Age	Body weight edema free	Period of observation	Blood pressure	Blood urea N per liter	Urea index $\frac{U}{B} \sqrt{\frac{V}{1.73}}$	Phosphate in 2 hours
		years	kg			gm		grams
44	S Ly	15	40	1923 January 21–February 18 February 20–February 22 March 4–March 27	158/100 165/102	0 28 0 17	13 6 13 6	5 1
45	J L	16	49	1924–1925 October 5–March 7 May 20–May 22	128/68 142/80	0 20 0 14	15 2 34 4	69 57
46	M McC	20	30	1925 February 19–May 19	146/90	0 17	20 5	53
47	E S	29	49	April 27–May 22	200/130	0 28	9 1	16
48	M H A	17	50	1921–1922 November 26–January 10 January 12–June 13	140/80	0 33 0 36	17 8 10 2	51 42
				1924 June 30–July 17		0 40	4 1	
		20		1925 January 19–February 17 February 27–March 17 March 22–April 21 April 22–May 12	160/130 155/130 150/130 130/100	0 92 0 66 0 84 1 14	4 5 3 5 1 5 1 3	Tract
49	S L	17	57	1921 November 1–November 5 November 6–November 24 November 27–December 11	188/122	0 31 0 20 0 12	23 0 9 6 12 5	16 20 31
				1922 February 1–April 12	140/70	0 29		29
				1923 December 26–December 29		0 49	8 0	8
50	M F	25	48	1923 January 7–January 21 January 23–February 13 February 15–March 8 March 11–May 27	180/130 155/105 145/95 160/110	0 78 0 54 0 53 0 55	6 2 5 0 5 9 5 1	17 20 26
51	C A	34	50	1924 October 13–December 19	180/104	0 72	5 2	5
52	E L	33	48	1923 December 2–December 19	220/120	0 47	6 3	Tract
				1924 January 13–January 27		0 37	5 2	Tract

	Protein intake per day	Ammonia excretion per day	Acid excretion per day	$\frac{\text{NH}_3}{\text{acid}}$	Blood $\text{NH}_3\text{-N}$ per 100 cc	Remarks
	gm	cc 0.1 N	cc 0.1 N	ratio	mg	
1	50	36	256	0.14		CaCl ₂ , 12 gm per day
	50	60	230	0.26		
	50	46	191	0.24		
2	55	62	311	0.20		
		24	100	0.24		
5	40-50	61	205	0.30	0.01	
7	50	48	139	0.35		
	30-60	132	321	0.41		
	60	90	300	0.30		
	20-40	51	122	0.42		
7	45	44	154	0.29		
	45	22	87	0.25		
		41	112	0.37	0.06	
		15	51	0.29		Died May 26, 1925
2	30	114	270	0.42		Acidosis, 60 gm. NaHCO ₃
	35	55	191	0.29		
8	30	107	176	0.61		Edema elimination
	50	74	190	0.39		
	40	76	340	0.22		Died June 1924
19	30-50	69	293	0.24		
	40-50	70	273	0.26		Edema elimination
	60-70	69	235	0.29		
	40-50	53	224	0.24		Died April 20, 1925
35	55	58	235	0.25		
34	30-40	53	176	0.30		
39	40-45	21	112	0.19		Died July, 1924

TABLE I

Number	Case	Age	Body weight edema free	Period of observation	Blood pressure	Blood urea N per liter	Urea index $\frac{U}{B} \sqrt{\frac{V}{IV}}$	Protein in 2 hours
		years	kg			gm		traces
53	E Sk	21	56	1921 November 18–December 11 December 18–February 23	200/150 160/110	0 60 1 28	4 5 2 6	Trace Trace
54	A S	27	47	1924 April 11–May 1	180/135	0 7–2 2		8
55	J M	28	55	1923 October 28–October 29	205/140	2 19		
56	M K	24	44	1923 October 5–October 6	200/105	1 58		
57	R S	31	44	1924 November 3–December 13 December 14–January 13	158/96 170/100	0 53 0 73	4 6 4 6	Trace Trace
				1925 January 14–February 17	182/112	0 82	3 8	Trace
58	J C	27	60	1923 October 25–November 13 November 15–December 20	170/117	0 42 0 62	9 1 6 6	27 11
				1924 March 20–April 14	195/130	2 87	2 7	
59	A Bl†	62	60	1925 April 30	200/140	0 72		<5
60	H F†	38	61	April 30	180/100	2 48		<10
61	V S	27	48	1921 October 24–October 30 October 31–November 1	206/126	1 55	3 9	
62	N H	52	53 4	1924 October 11–October 31	212/120	0 67	5 9	14

† These cases were obtained from the Presbyterian Hospital through the courtesy of Dr Atchley

Autopsies were performed on J D, M H A, S L, E L, E Sk, A S, J M, and V S, and the findings in each case confirmed the clinical diagnosis

Acid and ammonia excretion in nephritic acidosis As pointed out above, in normal individuals and in nephrosis the threat of acidosis induced by administration of calcium chloride leads to a somewhat

	Protein intake per day	Ammonia excretion per day	Acid excretion per day	NIH ₂ acid	Blood NIH ₂ -N per 100 cc	Remarks
	gm	cc 0.1 N	cc 0.1 N	mg. 10	mg	
	40-50	44	259	0 17		
	70-90	57	248	0 23		Died March 17, 1921
2	30-2	21	70	0 30		Died May 30 1924
	6-0	18	127	0 14		Died October 30, 1923
	30-0	9	135	0 07		Became incontinent Died October 11, 1923
	55	19	162	0 12		
	60	40	305	0 13		
	60	21	184	0 11		Died May 12 1925
	50-60	119	369	0 32		
	60	620	Neutral	∞		
	30-0	349	75	4 66		Died April 15, 1924
		748	Alkaline	∞	0 01	Uremia, edema, blood creatinine 4.2 mg in 100 cc.
		298	Alkaline	∞	0 02	Uremia, blood creatinine, 26.6 mg per 100 cc. Died May 21
	50	11	82	0 13		Malignant nephrosclerosis. Uremia, blood creatinine, 24.7 mg. in 100 cc.
		295	Alkaline	∞		Died November 1, 1921
	55	44	163	0 27		Malignant nephro-sclerosis

greater proportional increase in the excretion of ammonia than in that of acid, so that the ammonia-acid ratio increases. In contrast with this is the reaction of the kidney in chronic nephritis (7). Thus, in S. L., 44, whose ammonia-acid ratio was already low, the administration of calcium chloride led to no significant increase in the ammonia excretion or the ammonia-acid ratio. It is presumable, therefore,

TABLE 9
Acidosis and the ammonia-acid ratio in glomerulonephritis, Stage III
 Case A B, No 63 Age 8 Weight 18 kg

Date	Blood urea N per liter	Urea index	Ammonia excretion oz 0.1 N per day	Acid excretion per oz 0.1 N day	$\frac{\text{NH}_3}{\text{A}}$	Venous plasma			Alkali therapy	Remarks
						pH	CO ₂	Cl		
	gm				ratio		mM	mM		
1923										
November 27	0.91	5.2	56	108	0.52					
November 29			40	89	0.45					
December 2			44	68	0.50					
December 6			33	59	0.56					
December 9	1.57	1.9	21	57	0.37	7.05	8.9	89	7 gm NaHCO ₃	Convulsions and coma
December 10			21	54	0.39	7.46	12.6	74	6 gm NaHCO ₃	
December 13			21	52	0.40	7.20	11.6			
December 14			18	37	0.49	7.40	15.6	91	5 gm NaHCO ₃	
1924										
January 1			26	54	0.48					
January 6	1.64	1.9	18	43	0.42	7.35	13.4	93		
January 10			18	46	0.39					
January 15			10	19	0.53	7.25	15.1		0.5 gm NaHCO ₃ daily	
January 20			9	26	0.35					
January 24			10	28	0.36	7.29	14.5		1.0 gm NaHCO ₃ daily	
February 2			6	17	0.35					
February 10						7.30	16.4			
February 17	3.23	0.6	6	18	0.33	7.17	12.7	64		Died February 18

that the acidosis noted in the more severe forms of nephritis (A S, 54, and A B, 63) was due to inability of the organism to form more than small amounts of ammonia

The spontaneous occurrence of acidosis in a severe form is illustrated in the case of A B, 63, a boy of 8, who had had glomerulonephritis for 3 years and who was under observation for the last 3 months of his life (table 9) There was no history of edema and the

TABLE 10

The ammonia content of the blood in normal and nephritic subjects

Subject	Urea index	Ammonia excretion per day	Acid excretion per day	$\frac{\text{NH}_3}{\text{A}}$	Blood $\frac{\text{NH}_3-\text{N}}{\text{per 100 cc.}}$	Diagnosis
		cc 0.1 N	cc 0.1 N	ratio	mg	
L L	54 0	12 9*	12 4*	1 04	0 06	Normal
W N	44 2	23 5*	15 6*	1 51	0 10	Normal
G G	34 6	218	243	0 90	0	Nephrosis
M R.	28 2	134	116	1 16	0 01	Nephrosis
B F	31 5	312	166	1 88	0 02	Nephrosis
D G	73 8	232	181	1 28	0 01	Glomerulonephritis, Stage I
R. N	18 6	69	249	0 28	0 01	Glomerulonephritis, Stage III
P L	21 7	84	246	0 34	0 09	Glomerulonephritis, Stage III
M McC	20 5	61	205	0 30	0 01	Glomerulonephritis, Stage III
M H A.	1 5	41	112	0 37	0 06	Glomerulonephritis, Stage III
A. Bk.		748	Alkaline		0 01	Glomerulonephritis, Stage III
H. F		293	Alkaline		0 02	

* Excretion per hour

blood pressure was not raised There was retention of nitrogen at the time of admission and this rapidly increased He had convulsions which were associated with extreme uncompensated acidosis, this was partially controlled with bicarbonate and no further convulsions occurred With this persistent acidosis the excretion of ammonia and titratable acidity was small and steadily diminished As both were equally affected the ratio did not change Hence it appears that there is a possibility of only a moderately diminished ammonia-acid ratio

in severe forms of nephritis when the titratable acid excretion is also impaired, and acidosis develops. On the other hand, in spite of the low ammonia excretion, if the titratable acid remains at about the normal level, acidosis may be, and frequently is, absent.

Caution appears necessary in the use of calcium chloride, ammonium chloride, or any other acid forming diuretic, in the attempt to eliminate edema in patients with low ammonia-acid ratios, or with advanced glomerulonephritis. The kidneys may be unable to respond to the extra strain on the acid excreting mechanism, and acid intoxication may develop. It is desirable to follow the pH and bicarbonate of the serum closely during the use of such diuretics.

The ammonia of the blood Table 10 shows values for blood ammonia nitrogen in normal subjects and in different types of nephritis, including terminal uremia. The highest figure, 0.10 mg per 100 cc, occurred in a normal subject, and the lowest figures, 0 to 0.01, occurred in individuals with various forms of nephritis. Similar results have been reported by Rabinowitch (26) and Russell (28), who used the Nash and Benedict technique (21). In spite of the fact that there was a markedly diminished ammonia excretion in some of the cases, the blood ammonia showed no increase. These observations indicate that no retention of ammonia occurs comparable to that of other nitrogenous constituents. The diminished ammonia output is due not to retention but to diminished formation.

SUMMARY

In those forms of nephritis (*nephrosis* and *benign nephrosclerosis*) in which the urea excreting function is quantitatively unimpaired, both ammonia and titratable acid have been found to be excreted in normal amount and proportion. The usual ammonia-acid ratios in the urine in such cases exceed 1.

In *acute glomerulonephritis* a tendency to low ammonia-acid ratios was present in some cases, absent in others.

In those forms of nephritis with quantitatively impaired urea concentrating function (*chronic glomerulonephritis* and *some cases of acute glomerulonephritis*) there was regularly a low ammonia-acid ratio of 0.7 to 0.1 in the urine. It was due, as found by Henderson and Palmer,

to diminished ammonia output. The titratable acidity was relatively well maintained until the final stage of complete renal insufficiency.

The drop in the ammonia-acid ratio, was not always proportional to the fall in urea excreting power nor to the severity of the clinical condition. Cases with clinically mild chronic nephritis and urea concentration indices of 20 to 30 (instead of the usual normal above 40) may show ammonia-acid ratios as low (0.2 to 0.3) as other cases approaching uremia and with concentration indices below 5. A possible explanation of the failure of the ammonia-acid ratio to be proportionally depressed in cases with very low urea function is that in them the ability to excrete titratable acid as well as ammonia is reduced.

The ammonia content of the blood was as low or lower in those forms of nephritis with diminished ammonia output as in normal individuals. The fall in ammonia excretion in glomerulonephritis appears, therefore, due not to ammonia retention in the body, but to failure to form ammonia in proportions normally related to those of the acid metabolites. The concurrence of diminished ammonia formation with diminished kidney function is compatible with the conclusion of Nash and Benedict, that ammonia is formed in the kidney.

The determination of the ammonia-acid ratio in the urine is technically simple, and consistently normal values of the ratio in repeated determinations appear to exclude the possibility of serious renal impairment. The fall in the ratio, however, although present in all cases thus far observed with impaired urea concentrating power, does not regularly parallel the fall in the urea concentrating or excreting power. The decrease in urea function is more closely correlated with the clinical severity of the nephritis.

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THE EFFECT OF IODINE BY MOUTH ON THE REACTION TO INTRAVENOUS INJECTIONS OF THYROXIN

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(Received for publication November 2, 1925)

Repeated observations within recent years have shown that the oral administration of iodine will usually be followed by a striking reduction in the basal metabolism in patients with exophthalmic goiter. A histologic study of the thyroid glands which have been removed from these patients gives evidence that definite involutional changes follow the use of iodine which indicate an anatomic regression toward a normal or colloid resting state (1). It has, therefore, been assumed on very good evidence that the anatomic alteration of the gland, in part, at least explains the reduction in the basal metabolism and the associated improvement in the patient's condition. Another possible explanation of this phenomenon is that iodine may have a direct effect, in some unknown manner, on the circulating product of the thyroid gland whether the latter is an excessive or abnormal secretion or both. In order to obtain information bearing on the action of iodine on a pure excess of the secretion of the thyroid gland, the following experimental studies were carried out.

In brief, the plan of experimentation consisted in determining the effect of intravenous injections of thyroxin in rabbits as indicated by changes in the basal metabolism, pulse-rate, and body-weight. Following this the observations were repeated with similar intravenous injections of thyroxin, after the animals had been given iodine orally in the form of Lugol's solution. Under these conditions an opportunity was afforded to observe if iodine by mouth can alter, in any way, the animals' response to thyroxin.

METHODS

The animals used were healthy, male rabbits of various breeds. They were of different ages, although an attempt was made to secure about six months old rabbits, about one-half of these animals were

of this age During the course of the experiments, which usually extended over long periods, several animals developed "snuffles" As long as the animal's body temperature remained normal, this had no apparent effect on the body-weight, pulse rate, or basal metabolism as indicated by the control animals If the temperature rose above normal the experiment was immediately terminated The animals were kept in ordinary cages and fed a liberal diet consisting of oats and "greens" which was roughly measured A record was kept of all the food given which was not consumed

The heart rate was determined by means of an electrocardiograph tracing which was necessary as the normal rate of a rabbit varies between 200 and 250 per minute, and in some instances, following the thyroxin injections, it rose to over 400 beats per minute In order to keep the animals quiet during the pulse-rate determination it was necessary to restrain them on an animal board, which was not ideal as the heart rate was probably accelerated by these manipulations For comparative studies, however, this was satisfactory as indicated by the control animals in whom the heart rate remained very constant

The basal metabolism was calculated from the oxygen consumption, which was determined by means of a closed chamber apparatus comprised of an animal chamber with a water seal, a bell of 8 liters capacity suspended in water, an electric blower, and a soda-lime container for the removal of carbon dioxide The chamber and movable bell which recorded the contraction of air in the circuit and as much more of the apparatus as possible were enclosed in a water bath The chamber and bell were provided with thermometers, reading in hundredths of a degree centigrade, in order to record if the temperature in the circuit was constant throughout A small blower in the circuit was connected by a direct shaft to an electric motor which provided a ventilation of approximately 20 liters per minute Air leaks in the system gave very little difficulty after all the connections were once made tight Each day, however, a test of tightness was made on the apparatus before the experiment was begun To avoid any accumulation of carbon dioxide in the system the soda-lime was changed at suitable intervals, and as a further check the contained air, following an experiment, was forced through a solution of barium

hydroxide in order to detect the slightest trace of carbon dioxide. In no instance was there any evidence that the CO_2 was not being removed efficiently. The accuracy of the apparatus was determined by frequent alcohol check tests (2) and a variation averaging 5 per cent was found. As this error was constant and indicated that the observed oxygen consumption was always 5 per cent less than the theoretical, it was considered satisfactory for our purposes. As the animals were confined in a chamber in which it was impossible to see them move, it was necessary to use a suitable apparatus to record muscular movements. This was accomplished, in all but the first few experiments, by means of a recording pen which was attached to the movable spirometer bell. Each time the animals moved a graphic record was made on a revolving drum and thereby a permanent record of muscular movements was obtained. While the activity of different animals varied, it may be said that in general they were quiet, and it was usually possible to obtain satisfactory periods. In each instance in which the animals moved a significant amount, it has been noted and considered in the interpretation of the results. The animals were fed each day at about 5 p.m., and if any portion of the food remained in the cage the following morning its presence was noted and it was removed several hours before the metabolism determination. The procedure followed in determining the metabolism was to place the animal in the chamber for a preliminary period of one-half to one hour. After conditions were constant, and the animal quiet, the oxygen consumption was recorded for two fifteen minute periods. In almost all instances these periods agreed within 5 per cent. After having obtained the oxygen consumption, the figures were changed to calories, by the assumption of a respiratory quotient of 0.92, and by reference to the table of Zuntz and Schumburg (3) which gives the calorific value of a liter of oxygen for various non-protein respiratory quotients. The results were then expressed in per cent of normal by use of the average normal values of heat production per kilo in rabbits given by E. Voit (4). Although some question may arise concerning correctness of the assumed respiratory quotients and normal values of heat production for rabbits which were used, this has no bearing on the present use of them, as the observations are merely comparative.

It would have been possible to utilize the actual oxygen consumption, expressed in cubic centimeters with as satisfactory results. The thyroxin used was Squibbs crystals of the highest grade. It was given in doses of 1 mg. after being put in solution in distilled water by rendering it slightly alkaline with dilute sodium hydroxide. The Lugol's solution (Liquor Iodii Composition) was given in doses of 0.1 cc. by mouth.

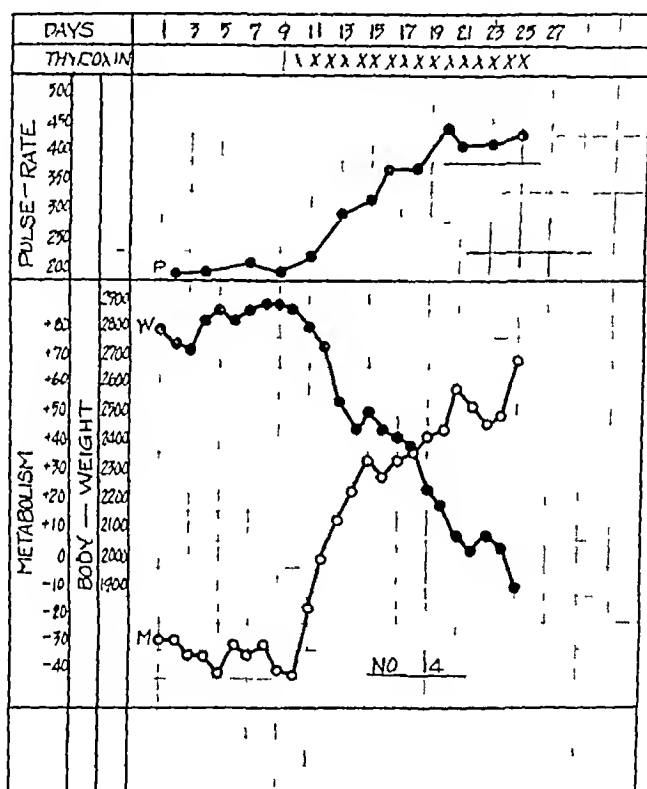


CHART 1 EXPERIMENT 14 THE EFFECT OF 1 MG. OF THYROXIN INTRAVENOUSLY FOR SIXTEEN CONSECUTIVE DAYS

P, pulse-rate recorded in beats per minute, W, body-weight in grams, M, metabolism in per cent of normal, X, 1 mg. of thyroxin intravenously

THE EFFECT OF THE INTRAVENOUS INJECTION OF THYROXIN IN RABBITS

Chart 1 (experiment 14) illustrates the effect of the administration of thyroxin over a period of sixteen days. After a control period of nine days in which the metabolism and body-weight was recorded

daily, and the pulse rate determined on four days of the period, the thyroxin injections were begun in doses of 1 mg intravenously and continued for sixteen consecutive days as indicated by the cross marks at the chart. It will be noted that during the control period the pulse-rate remained very constant, the range being between 210 and 230 per minute. The body-weight was likewise fairly constant, although there was a slight tendency for the animal to gain weight

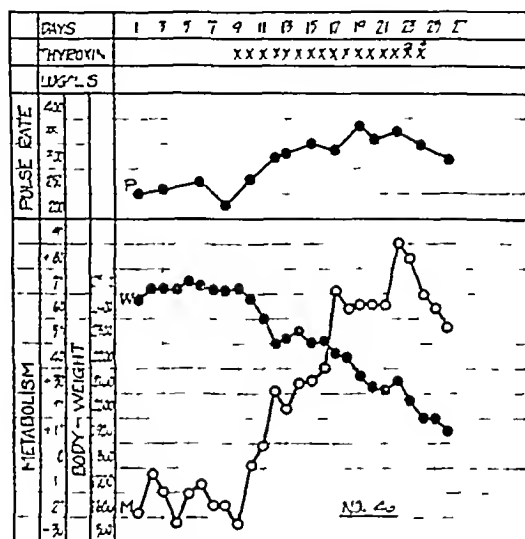


CHART 2 EXPERIMENT 46 THE EFFECT OF REPEATED MILLIGRAM DOSES OF THYROIDIN INTRAVENOUSLY ON THE PULSE-RATE, BODY-WEIGHT AND METABOLISM

As there was an immediate drop in weight following the use of the thyroxin, this made the effect even more convincing. During the control period the basal metabolism varied as much as 12 per cent, although the variations from one day to the next was very much less, and there was no constant tendency for the metabolism curve to proceed upward or downward. Following the injection of thyroxin there was an effect, slight but definite, within twenty-four hours as indicated by an accelerated pulse-rate, a decrease in body-weight, and an increased metabolism. As the drug was continued there

was a remarkable symmetrical rise in the pulse-rate which was paralleled by the metabolism. After the thyroxin had been injected for sixteen consecutive days, the pulse-rate was 450 per minute, and the metabolism had increased from -39 to $+71$. Coincident with these changes the body-weight diminished from 2900 gm to 1925 gm, which represents a loss of 975 gm or approximately 35 per cent of the original body-weight. Although the animal's food consumption was only measured roughly, it was apparent that a normal amount

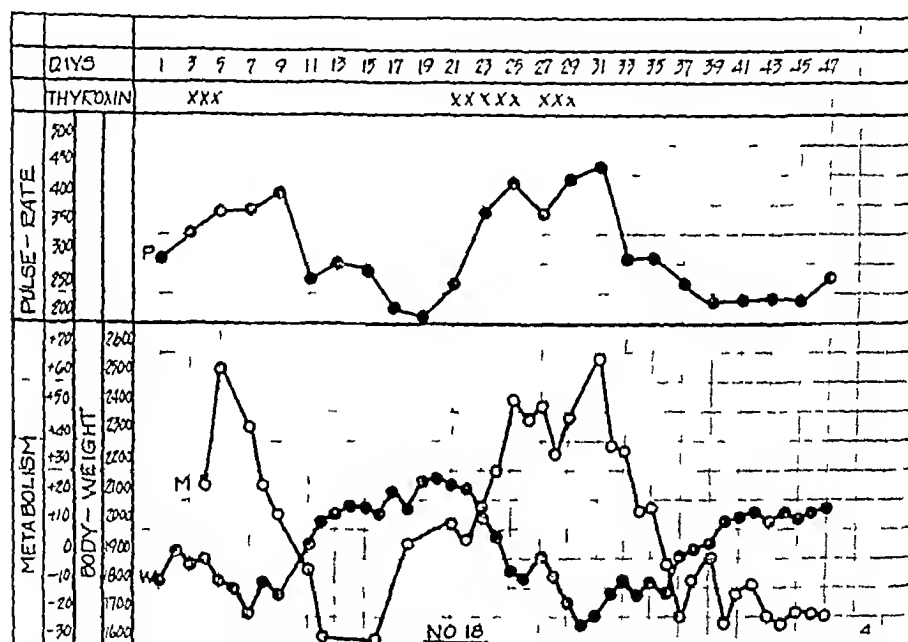


CHART 3 EXPERIMENT 18 THE EFFECT OF THYROXIN WHICH WAS GIVEN IN MILLIGRAM DOSES FOR THREE CONSECUTIVE DAYS, THEN OMITTED FOR FIFTEEN DAYS, AND RESUMED FOR EIGHT DOSES

was consumed during the major portion of the experiment, although much less was eaten during the last few days of observation. It was obvious, however, that the chief cause of the loss of weight was due to the elevated metabolism. The experiment was terminated on the twenty-fifth day as it was clear that the characteristic effects of thyroxin had been demonstrated. These results were duplicated in experiment 46 (chart 2) in which 1 mg of thyroxin was given for fourteen consecutive days after a satisfactory control period of eight

days The thyroxin was successfully given on twelve days On the succeeding two days, attempts were made to administer the drug but probably only a small amount was actually injected as the ear veins were in poor condition Chart 3 (experiment 18) shows the characteristic response to thyroxin, first, following its injection on three consecutive days, and again after an interval of sixteen days, at which time the drug was given in milligram doses for eight days The striking increase in pulse-rate and basal metabolism when considered with the rather abrupt drop in body-weight, which follows shortly after the injection of thyroxin intravenously, appears to be entirely convincing that this drug is responsible for these changes and indicates that a rabbit will respond with characteristic alterations in pulse-rate, basal metabolism and body-weight after thyroxin is administered in doses similar to those used in the above experiments

THE EFFECT OF THE SIMULTANEOUS ADMINISTRATION OF THYROXIN AND IODINE

Experiment 14 (chart 4) shows observations on an animal over a period of forty-nine days This chart shows, first, the customary changes in the pulse-rate, basal metabolism and body-weight which are associated with the intravenous injections of thyroxin on three consecutive days After this response had subsided, Lugol's solution, by mouth, was begun in daily doses of 0.1 cc, well diluted with water, and continued throughout the experiment After this dosage had continued for eight days, thyroxin was also given daily in milligram doses for eight days, the animal received, therefore, 0.1 cc of Lugol's solution and 1 mg of thyroxin daily for this period A comparison with the alterations in pulse-rate, basal metabolism and body-weight in the two periods during which the thyroxin was administered, shows that while the changes in the second period may be slightly greater than in the first, yet at this time more than twice as much thyroxin was given, and the animal's response was in every way similar to that elicited in an animal by the injection of a similar amount of thyroxin and to which Lugol's solution had not been given (see chart 3) It is apparent from this chart that the animal gained considerably in body-weight during the forty-nine day period of study This is most readily explained by the observation that con-

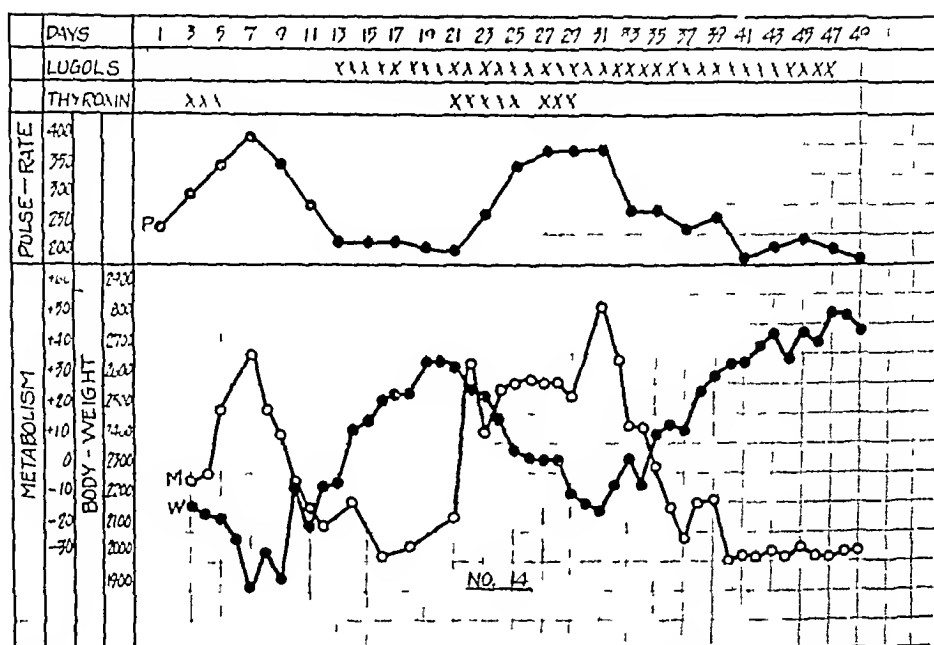


CHART 4 EXPERIMENT 14 ILLUSTRATING THAT IODINE BY MOUTH DOES NOT EFFECT THE RESPONSE TO THYROXIN

At the top of the chart, X following Lugol's, indicates that 0.1 cc was given by mouth on that day

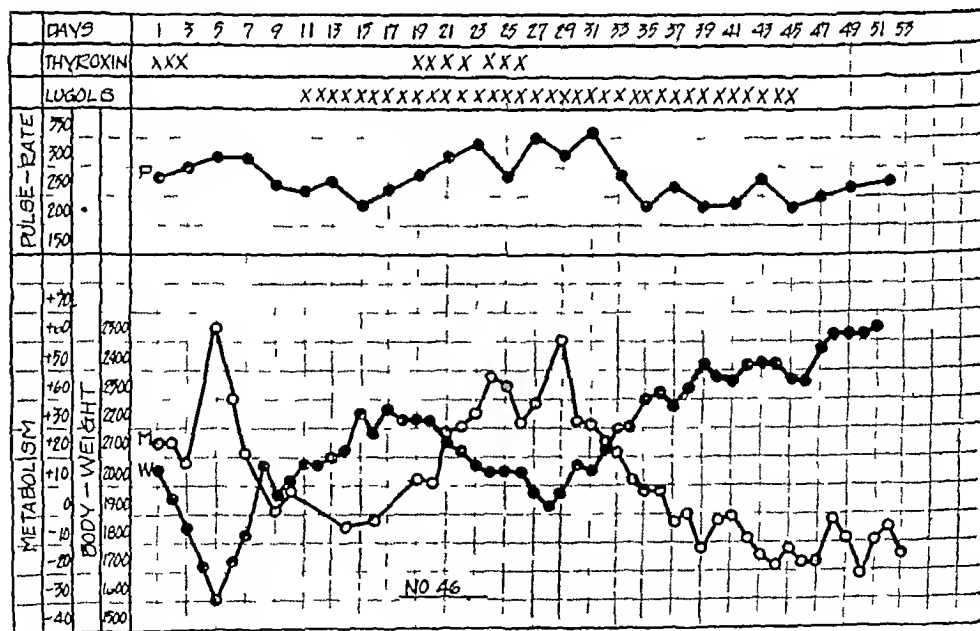


CHART 5 EXPERIMENT 46 A DUPLICATE OF THE RESULTS SHOWN IN CHART 4

trol animals in the laboratory gained a similar amount of weight which indicated that the environment, in which all of the animals were kept, permitted this. Experiment 46 (chart 5) is a duplicate of experiment 14, and confirms in every respect the changes which had previously been observed. Additional studies were made which amply confirmed these experiments and indicated that observations in this direction would be a needless duplication of results.

DISCUSSION

From these experiments it is apparent that striking changes in the pulse-rate, basal metabolism and body-weight may be produced in rabbits by the intravenous injection of thyroxin. These changes are characteristic as they appear regularly following the injection of the drug, and they are similar to changes observed in patients with exophthalmic goiter although it can not be stated that the typical syndrome of the disease has been produced in animals. Additional observations indicate that the saturation of an animal with iodine by mouth in the form of Lugol's solution, had no effect on the anticipated response to injections of thyroxin. If thyroxin is the active principle of the thyroid gland which is now generally accepted and it is assumed that the syndrome of exophthalmic goiter results from a pure excess of the substance, then it seems clear, from these experiments, that iodine has no effect on the circulating thyroxin, but must exert its influence on the rate of production of this substance in the gland itself. In addition, judging from a few unreported observations made in this laboratory, there is no evidence which suggests that iodine by mouth, at least in normal individuals diminishes the normal rate of secretion of the thyroid gland, as the administration of iodine in man is not followed by a decrease in the basal metabolism. The experiments which are reported in this article are not incompatible with Plummer's theory (5) that the symptoms of exophthalmic goiter are due to an altered secretion of the thyroid gland and that this abnormal secretion may become more nearly normal following the administration of iodine. They furnish no support to it, however, except that further evidence is produced which is in accord with the theory that exophthalmic goiter is not due to a pure excess of thyroid secretion.

CONCLUSIONS

1 The intravenous injection of thyroxin in rabbits produces a characteristic increase in the pulse-rate, basal metabolism and a decrease in the body-weight

2 The administration of Lugol's solution has no effect in altering the characteristic response, in rabbits, to injections of thyroxin.

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THE EFFECT OF A STANDARD EXERCISE UPON THE OXYGEN CONSUMPTION OF NORMAL, OVERWEIGHT, AND CHRONICALLY ILL INDIVIDUALS*

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(Received for publication January 4, 1926)

INTRODUCTION

This work was originally undertaken eighteen months ago, with the thought that possibly a study of the oxygen consumption during a standard exercise and the length of time required for the subject to pay off his oxygen debt might be of some value in appraising the fitness of an individual. We were disappointed to find that normal individuals varied widely in their percentage of oxygen consumption during exercise, according to their surface area. However, in duplicating the tests on the first subjects studied, we found that the response to exercise was constant in the individual, indicating that the test was accurate despite the wide variation that occurred in different subjects. Because of this individual constancy, it was decided to extend the study to include individuals who were overweight and those who had miscellaneous chronic diseases.

The literature upon the effect of work upon respiratory metabolism has been so recently reviewed by Boothby and Sandiford (4) that it seems unnecessary to include an extensive consideration of it here, particularly when so little of it has a bearing on the phase of the problem to which this work pertains.

Smith (8) in an extensive study upon walking finds that in eight subjects the increase in energy expended in horizontal walking from standing varied from 0.454 to 0.618 gram-calorie for each horizontal kilogram-meter, and that variations in speed under 80 meters per minute had no effect upon the results, but above that point the expenditure of energy increased proportionately with the speed.

*Presented in abstract before the joint meeting of The Federation of American Societies for Experimental Biology, at Cleveland, Ohio, December 30, 1925.

Strouse, Wang and Owen (9) report a résumé of a study of the mechanical efficiency, as measured by a bicycle ergometer, on thirteen obese, ten normal, and six thin subjects. Their figures show that the obese spend more energy on a given piece of work than the normal and that the thin people are the most efficient of the three groups. Peabody, Sturgis, Barker and Reed (7) compared the oxygen consumption and pulmonary ventilation of eleven ambulatory cardiac patients with normal subjects, during standing, and walking sixty steps. The same relative changes in the oxygen consumption and pulmonary ventilation were found in the two groups.

In the monograph by Hill and Lupton (6) upon Muscular Exercise, Lactic Acid, and Supply of Oxygen, they state, that in a moderate exercise the respiratory quotient should remain at unity, because the protein buffers of the muscle should neutralize all the lactic acid formed, so that little CO_2 should be driven out. In their experiments they found that after a moderate exercise the oxygen intake would return to its resting level in six to eight minutes. Their observations, we imagine, were made with atmospheric air, while ours were made with pure oxygen so the results may not be comparable.

Benedict and Benedict (2) studied the effect of minor muscular movements upon the metabolism and found that some of them had a definite influence upon the consumption of oxygen,—thus, fifteen movements of the arm per minute produced an increase in the metabolism of about 11 per cent, and three movements of the leg per minute caused an increase of about 13 per cent in the metabolism.

Boothby and Sandiford, whose work will be referred to later on, have shown, in a series of fourteen patients with hyperthyroidism, that they require nearly twice as much energy to perform a given piece of work as do normal persons or patients with other diseases.

METHOD OF STUDY

The subjects were all studied in the post-absorptive state, not having had food for twelve to fifteen hours. The Portable Benedict (1) apparatus was used for all the determinations. The surface area formula of DuBois and DuBois (5) was used in calculating both the basal and exercise metabolic rates. The formulas proposed by West (10) were used in the calculation of the vital capacity in percentages, the height formula being used for those who were obese or overweight, and the surface area formula for all the others.

In the choice of a satisfactory exercise with which the Benedict Portable apparatus could be used, we thought it best to select some activity which would be comparable to walking so that it could be made as nearly automatic as possible and so that the relation of the amount of work done to the individual would be as near a constant as possible. The subject lay on his back and was required to lift his lower extremities rhythmically and alternately, so as to touch with his toes a string which was placed a given distance, corresponding to about one pace, from the plane on which he was lying.

TABLE I
Normal subjects (arranged in order of age)

Number	Date	Sex	Weight	Height	Age	Vital capacity	Basal O ₂ per minute	Exercise O ₂ per minute	B.M.R.*	E.M.R.†	Basal exercise ratio	Recovery period
			kg	cm		per cent S.A.	cc	cc	per cent	per cent		minutes
3	12-10-24	M	60.7	168	18	88	302	512	+12	+99	1.177	3
28	12-11-24	M	59.8	167.5	25	98	243	456	-3	+92	1.198	3
	1-10-25		60.4	167.5	25	103	201	390	-19	+62	1.200	3
23	12-4-24	F	58	162	19	105	223	535	-3	+144	1.253	6
	2-13-25		59.6	162	19	104	205	496	-13	+118	1.256	6
24	2-5-25	M	56.7	175	26	117	218	446	-15	+84	1.216	4
	2-12-25		58.2	175	26	117	216	441	-17	+77	1.213	4
21	12-6-24	F	55.6	169	28	95	225	503	-3	+127	1.234	
	2-14-25		54.5	169	28	96	220	520	-3	+137	1.244	4
56	12-9-25	F	53.1	168	27	89	230	500	-1	+121	1.223	3
10	2-10-25	M	51.7	164	16	96	258	663	+1	+170	1.267	4
	2-14-25		52.3	164	16	97	265	641	+3	+161	1.253	5

* B.M.R.—basal metabolic rate expressed in per cent above or below 100

† E.M.R.—exercise metabolic rate expressed in per cent above or below 100

The subject's basal metabolism was first determined after a period of rest, usually at least thirty minutes, allowing a sufficient time for the pulse rate and blood pressure to become stabilized. No subjects having any degree of fever were studied. The apparatus was tested for leakage before each experiment by placing a 100 gram weight on top of the spirometer which was filled with oxygen, and checking the reading at the end of six minutes. We required that the oxygen consumed in six minutes should check another period within 20 cc. After the

TABLE 2
Obese and overweight subjects (arranged in order of weight)

Number	Date	Sex	Weight	Height	Age	Vital capacity	Basal O ₂ per minute	Exercise O ₂ per minute	B M R	E M R	Basal exercise ratio	Recovery period
			kg	cm		per cent height	cc	cc	per cent	per cent		min utes
32*	6-20-25	F	151 5	160 5	34	75	352	692	+11	+128	1 2 05	3
	6-30-25		147 7	160 5	34	75	335	688	+6	+128	1 2 15	3
47	4- 7-25	F	137 5	164	54	82	268	675	-14	+125	1 2 61	4
34	3- 2-25	F	130 2	166	16	124	292	657	-20	+97	1 2 46	4
2†	12-10-24	F	119	172	48	73	248	430	-20	+45	1 1 81	
15	2-27-25	F	116 6	159	30	81	263	553	-16	+92	1 2 29	4
54	11-27-25	F	109 1	157	11	83	252	602	-27	+83	1 2 50	4
17	2-18-25	F	106 2	156	46	99	272	520	+1	+98	1 1 97	3
48*	5-22-25	F	88	161	85	84	232	547	-14	+116	1 2 51	3
22	1-30-25	M	85 3	167	33	91	268	608	-8	+115	1 2 34	4
	2-14-25		85	167	33	91	263	597	-11	+112	1 2 38	5
40	12-19-24	F	84	147	34	85	272	820	+10	+255	1 3 23	6
18	22-17-25	F	82 6	158	27	107	240	622	-8	+151	1 2 73	4
31*	4-11-25	M	81 7	166	52	84	235	672	-13	+166	1 3 05	3
44*	3-10-25	F	80 7	149	64	74	232	480	+1	+116	1 2 18	3
50*	3-28-25	F	80 5	163	29	107	242	622	-7	+137	1 2 55	4
13	12- 4-24	F	78 5	163 5	34	106	245	455	-1	+90	1 1 91	
	2-11-25		77 6	163 5	34	113	240	467	-7	+89	1 2 03	4
25	12- 9-25	F	72 8	163 3	28	86	242	555	-5	+127	1 2 39	7
53†	11-23-25	F	69 6	167 5	34	110	195	447	-21	+184	1 3 56	6
55	12- 5-25	F	69 1	157 5	12	86	270	550	-5	+113	1 2 24	3
38*	12- 6-24	F	68 7	163	17	86	252	550	-7	+112	1 2 28	5
27	12- 2-24	F	67	163	24	98	240	692	+1	+149	1 2 47	
	2- 4-25		67	163	24	101	242	585	+1	+159	1 2 56	4
26	12- 3-24	F	60 7	164	24	85	232	475	+1	+114	1 2 12	
	2- 3-25		61 5	164	24	87	237	467	+3	+111	1 2 05	6

* Diabetic

† Anemic type of obesity with diabetes Patient fatigues so that only one six-minute exercise observation could be made

‡ "Breeches" type of obesity

basal metabolism was satisfactorily determined, the oxygen consumption during exercise was determined in the following way A string was placed across the foot of the cot 30 inches above the surface so that the subject could easily touch it with his toes as his lower extremity swung through the arc A metronome was placed

near the foot of the cot and set so that it clicked sixty-six times per minute. The subject was then instructed to touch the string with the toe at the first beat of the metronome and to touch the bed with the heel at the second beat, alternating the lower extremities. In other words, each lower extremity was lifted so that the toe touched the string about seventeen times per minute. It appeared that this exercise, to the well-beaten time, became, after a moment's practise, very rhyth-

TABLE 3
Diabetic subjects (arranged in order of weight)

Number	Date	Sex	Weight	Height	Age	Vital capacity	Basal O ₂ per minute	Exercise O ₂ per minute	B M R	E M R	Basal exercise ratio	Recovery period
			kg	cm		per cent S.A.	cc	cc	per cent	per cent		minutes
32	6-20-25	F	151.5	160.5	34	75	352	692	+11	+128	1.2.05	3
	6-30-25		147.7	160.5	34	75	335	688	+6	+128	1.2.15	3
2	12-10-24	F	119	172	48	73	248	430	-20	+45	1.1.81	
48	5-22-25	F	88	161	35	84	232	547	-14	+116	1.2.51	3
31	4-11-25	M	81.7	166	52	84	235	672	-13	+166	1.3.05	3
44	3-10-25	F	80.7	149	64	74	232	480	+1	+116	1.2.18	3
50	3-28-25	F	80.5	163	29	107	242	622	-7	+137	1.2.55	4
43	3-19-25	F	73	164.5	37	81	268	677	+2	+179	1.2.74	3
51	3-31-25	M	72.7	169	19	99	252	550	-13	+99	1.2.40	3
38	12-6-24	F	68.7	163	17	86	252	550	-7	+112	1.2.28	5
12	1-23-25	F	66.2	170	57	85	198	417	-14	+88	1.2.19	4
36	3-6-25	M	61	170.5	29	109	215	438	-15	+76	1.2.07	3
41	12-5-24	M	60.8	164	65	68	248	422	+7	+91	1.1.77	4
35	3-2-25	M	60.7	157.5	31	109	232	528	-4	+128	1.2.38	3
52	3-24-25	M	58.9	161.5	38	91	228	658	-6	+181	1.2.99	3
46	3-5-25	F	57.9	160	53	100	227	383	+6	+88	1.1.77	3
20	2-21-25	M	57.7	176	46	103	215	453	-15	+89	1.2.22	4
49	4-8-25	F	57.2	150.5	58	82	198	478	-1	+149	1.2.51	3
45	3-4-25	M	55.7	177	26	104	215	520	-16	+112	1.2.52	3
16	2-28-25	F	55.5	161	51	96	210	430	+2	+118	1.2.13	2
42	3-18-25	M	48.2	161.5	26	110	275	763	+23	+253	1.2.87	4
	12-10-25		52.5	164.5	26	95	252	670	+14	+218	1.2.78	4

mical and easy for the large majority of the subjects. As soon as the subject got well into the swing of this exercise, the spirometer reading was taken by the Emmes method, and it was taken again at the end of the six minutes when the subject was instructed to stop the exercise. A three minute rest period was then given with the subject detached from the mouthpiece. Another six-minute period was then run in the same manner at the end of which, however, the subject was not detached from the apparatus but allowed to breathe oxygen after the exercise.

was stopped. The readings were then taken every minute until the reading for one minute corresponded within a few cubic centimeters of the basal oxygen consumption for one minute.

The pulse was counted every five minutes during the rest period before the basal metabolism was taken, and before, once during, and after each exercise period.

The calculations were all based upon a universal respiratory quotient of 0.82 during rest, and of unity during exercise. It is probable that the exact respiratory quotient lies somewhere between those two figures, and it certainly is improbable, judging from the work of Hill, and Benedict and Cathcart (3), that such a moderate exercise would give a respiratory quotient over unity. In any event the difference in exercise metabolism, whether figured with a respiratory quotient of 0.82 or unity, would usually be less than 5 per cent, which is negligible.

The data obtained are presented in tabular form.

DISCUSSION

Seventy-one observations on the metabolism during exercise were made on fifty-six subjects and in fifty-five of them the periods checked within 160 cc, in the remaining sixteen, the difference was slightly more than this. However, of the fifty-five who did check within 160 cc, many checked within 40 cc and a few exactly.

Many of the subjects complained of slight fatigue following both of the periods of exercise. However, very few experienced moderate fatigue, and only one, subject 2, an individual with diabetes and obesity, had excessive fatigue so that she did not care to go through the second exercise period. It is noteworthy, that with this subject we obtained about the lowest percentage increase in metabolism during exercise. All of the other subjects had sufficiently recovered by the end of the three-minute rest period following the first exercise to go through the second period willingly.

Of course, it is obvious that the amount of work done by each individual is different and that it is impossible to quantitate the amount of this work. However, the length and weight of the lower extremity bear a more or less constant ratio to the length and weight of the trunk so that the amount of energy expended is relative to the individual. The results of the increase in metabolism produced by the exercise must then be expressed in percentages above the normal basal metabolic rate, which has been computed by the surface area formula, so that they will be comparable. Had we used a measurable

TABLE 4
Miscellaneous cases (arranged in order of weight)

Num ber	Date	Sex	Weight kg	Height cm	Age	Vital capac- ity percent %A	Basal O ₂ per minute cc	Pre- cho O ₂ per minute cc	N M R percent	L M R percent	Basal exercise ratio	Recov ery period minutes	Diagnosis
39	12-20-21	M	77.3	180	18	82	252	568	-11	+106	1 2 32	1	Auricular fibrillation
14	2-11-25	M	71.2	176	26	116	205	563	-30	+100	1 2 86	1	Idiopathic decrease in B M R
	1-27-25		75.1	176	26	113	200	546	-29	+102	1 2 86	5	
8	1-11-25	I	71.1	168	35	91	202	472	-19	+96	1 2 11	1	Post-operative thyroid
5	12-12-21	M	69.3	163	33	92	305	668	+15	+164	1 2 29	6	Chronic endocarditis
9	1-21-25	M	67.7	177	25	111	207	375	-25	+14	1 1 92	3	Psychoneurosis
29	6-11-25	I	63.3	161	49	106	182	188	-19	+127	1 2 80	1	Myocardema
7	12-29-21	M	58.7	175.5	39	88	302	645	+16	+199	1 2 50	7	Thyro toxic adenomata
6	12-30-25	M	57.7	176.5	16	51	200	462	-31	+68	1 2 13	1	Mitral stenosis
19	12-16-21	M	55.7	173.5	32	95	183	583	-28	+139	1 3 32	8	Auricular fibrillation, heart block
37	12- 9-21	I	49.5	159.5	17	91	218	670	-1	+208	1 3 20	6	Mitral stenosis
30	6-16-25	M	48.1	166	15	68	293	625	+9	+133	1 2 14	3	Aortic regurgitation
33	1-22-25	I	45.7	165	19	110	232	752	+9	+269	1 3 38	6	Hyperthyroidism
	1-28-25		47.6	165	19	103	228	605	+8	+198	1 2 76	6	
	3-19-25		48.3	165	19	102	223	572	-1	+168	1 2 71	4	
11	12- 3-21	M	48.5	161	31	76	232	650	+1	+205	1 2 93		Kypho scoliosis
	2-10-25		47.3	161	31	74	233	658	+5	+209	1 2 94	3	Pigeon breast
1	12-10-24	I	46.1	157	26	93	228	612	+11	+212	1 2 81	5	Petit mal attacks
1	12- 9-21	I	41	164.5	15	85	202	685	+3	+213	1 3 01	6	Mitral stenosis

amount of work such as lifting a weight through a specified distance there would also have been inconstant factors which are unmeasurable such as the relation of the individual to the amount of work, and the energy consumed by the part of the body which was selected to move that weight

In going over the data that have been presented in the tables it is evident that the large variation in oxygen consumption, both absolute and relative, during this standard exercise, in different individuals, makes any analysis or appraisal extremely difficult. This variation is, no doubt, attributable to many indeterminate factors such as possibly, fitness of the individual, temperament, and unequal ratios between the weight and length of the lower extremity to the weight and length of the trunk. Nevertheless, we feel that the energy expended in performing his exercise would be, in general, comparable to the work done while the subject was walking at the same rate and with the same pace.

It is quite evident from the tables, that the rate of the basal metabolism usually predetermines to some extent the rate of the oxygen consumption during exercise, those with a low basal metabolism, even though they were not definitely abnormal, showed a tendency to consume less oxygen during exercise, and on the other hand, those having even a slight increase in the basal metabolism usually consumed relatively more oxygen during activity. There are, however, a few notable exceptions to this observation. For instance subject 19, who had chronic myocarditis, auricular fibrillation, and at times, heart block, without cardiac failure, had a basal metabolism of minus 28 per cent while his metabolism during exercise was plus 139 per cent and his basal exercise ratio was 1.332, it is of interest, however, to note that he required eight minutes to return after exercise to his basal metabolic rate. Subject 53, who had recently developed a "breeches" type of obesity, had a basal metabolism of minus 21 per cent, but an exercise metabolism of plus 184 per cent, this probably is to be expected because of the relatively increased weight of the lower extremities. Furthermore, the accuracy of this observation is borne out by the fact that in the fourteen subjects on whom we were able to get duplicate observations, and on one, subject 33, on whom triplicate observations were made, the oxygen consumption during exercise

checked very well with the previous determination if the basal metabolism did. Four subjects, 23, 33, 42 and 28, had a lower oxygen consumption at the second test which corresponded proportionately to the lower basal metabolism which they manifested at the same time. Subject 33 showed the clinical manifestations of hyperthyroidism without any marked increase in the basal metabolic rate. Her oxygen consumptions during exercise were plus 269 and plus 198

TABLE 5

Condensed data on fourteen subjects upon whom duplicate observations were made

Subject	First test		Second test		Basal-exercise ratio	
	B.M.R.	E.M.R.	B.M.R.	E.M.R.	First test	Second test
	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>		
28	97	192	81	162	1 1 97	1 2 0*
33	99	268	108	298	1 2 74	1 2 76
23	97	244	85	218	1 2 53	1 2 56
42	123	353	114	318	1 2 87	1 2 78
24	85	184	87	177	1 2 16	1 2 13
21	97	227	97	237	1 2 34	1 2 44
10	101	270	103	261	1 2 67	1 2 53
32	111	228	106	228	1 2 05	1 2 15
22	92	215	89	212	1 2 34	1 2 38
13	99	190	83	189	1 1 91	1 2 03
27	101	249	101	259	1 2 47	1 2 56
26	101	214	103	211	1 2 12	1 2 05
14	70	200	71	202	1 2 86	1 2 86
11	104	305	105	309	1 2 93	1 2 94

* Example. In the ratio 97 192 81 162 the product of the means (15,552) very nearly equals the product of the extremes (15,714). The ratios are determined by dividing the actual exercise rate by the actual basal rate, thus $\frac{192}{97}$ equals 1.978, therefore the ratio is 1 1 978.

per cent respectively, with a six-day period of rest intervening. About six weeks following a thyroidectomy, her basal metabolic rate had dropped to minus 1 per cent and her exercise rate had dropped to plus 169 per cent. The condensed data from the fourteen subjects upon whom duplicate observations were made are presented in table 5. The first four subjects were the only ones who showed any essential change in their basal and exercise metabolism. It will be noted that in each case the product of the means almost exactly corresponds to

the product of the extremes, and that the ratios on the first and second tests were about equal, even though the results of the metabolism determinations varied

Upon critical examination of the results reported on the cost of work in a series of five normal subjects and eighteen patients with miscellaneous diseases studied by Boothby and Sandiford, it will be seen that those having basal metabolic rates lower than the normal standard required fewer calories for each kilogram-meter, and on the other hand, those having higher basal metabolic rates required more, with the prominent exception of the three patients with leukaemia who had a definite increase in metabolism but a normal response to exercise, and one case of myxoedema whose metabolism was low during rest but slightly elevated during activity, as was observed in our case. These authors, also, made repeated observations on one case of exophthalmic goitre before and after thyroidectomy, during basal conditions and while walking on a treadmill taking 24 to 30 steps per minute. Upon analysis of these data it is remarkable that the basal exercise ratio remained at about 1.2 during the many observations even though the basal metabolic rate varied from plus 59 per cent before operation to plus 7 per cent afterwards, and the metabolic rate during exercise varied from plus 221 per cent before to plus 108 per cent sixteen days afterwards.

In analyzing the basal-exercise ratios of our series, it was found that the average of the ratios for the twenty subjects who were below or of normal weight was 1.231 and the average of the twenty-one who were overweight or obese was nearly identical—1.245. The basal-exercise ratios of the fourteen subjects on whom repeated experiments were done agreed within a small decimal, even though the basal metabolic rate had changed appreciably as it had in four of the subjects. In subject 33, a case of mild hyperthyroidism, however, the ratios of the first and second tests did not agree, but the results of the second and third readings did. In this case there was a period of six days rest between the first and second tests and a thyroidectomy between the second and third. In view of these findings, and the wide variation that occurred in the basal exercise ratios of all subjects, it is at least suggested that all people have a rather individual response

to effort which tends to bear a more or less constant relation to their basal metabolic rate

It has been the experience of one of us (B D B) and of many other observers, that the caloric value of the diet does not play as important a rôle in the gain or loss of weight as one might expect, thus some patients fail to gain weight on high diets, and, on the other hand, some obese people do not respond to surprisingly low diets, even though they have been under strict hospital supervision. The present study of the response to exercise does not clarify this phenomenon. Subject 31, for instance, an active man with diabetes, who received a diet of 1900 calories and 20 units of insulin daily for the past three years, during which time he gained 20 kilograms in weight, had an exercise metabolism of plus 166 per cent and a basal-exercise ratio of 1.305. Our observations appear to be further proof of the futility of estimating a patient's diet by adding a certain percentage to his basal calories to provide for his activities.

It is unfortunate that we have not a larger group of individuals who could be classified as strictly normal with regard to their weight, height, basal metabolism and general health—only seven can be included in this group. It will be noted that the highest difference in the percentages of oxygen consumption for these subjects varied from plus 84 to plus 170 per cent. However, in the obese and overweight group, there are six individuals who in our opinion, are classified as overweight. The response to the exercise in this small group was fairly constant as will be noted from table 2, with the exception of subject 53 whose accumulation of fat was almost entirely confined to the lower extremities.

Of the five diabetic patients who were definitely thin (52, 20, 45, 16 and 42) there is a variation in the exercise metabolism from plus 89 per cent to plus 253 per cent, which is somewhat relative, however, to the basal metabolic rate. We were particularly interested in the findings of this group because, with the exception of subject 20, their diabetes had been under control for some time and they had been on rather high diets. They had, for some reason, failed to gain weight in proportion to the caloric value of their diets as other patients had done. This apparently could not be explained on the basis of a high

oxygen consumption, especially when we consider subject 42 who had the highest metabolism and who had gained the most weight, increasing from 48.2 to 52.5 kg in seven months, with a slight decrease in both his basal and exercise metabolic rates

The average rate of the metabolism determinations during exercise of the twenty diabetic and normal subjects who were of normal weight was 121.9 per cent and the average rate of the twenty-one obese and overweight subjects, both diabetic and non-diabetic, was 124.4 per cent. It then does not appear that weight is a prominent factor in bringing about variations in oxygen consumption during exercise in fact obese individuals seem to use about as much energy in proportion to their surface area as do normal individuals

The subjects composing the obese and overweight group do not appear to have such wide variations in their response to exercise as was demonstrated in the other groups. Subject 40, however, is an exception. This patient was the only one who because of her short lower extremities had difficulty in reaching the string and experienced considerable fatigue as the result of the effort. Her oxygen consumption during exercise was considerably increased as was her recovery period after the exercise. It is quite possible that in this case the slight increase in the basal metabolism (plus 10 per cent) together with the extra effort might have contributed somewhat to the high exercise metabolic rate. It is not our belief, however, that differences in the ease with which the string could be reached by the toes was a common cause of variations in the response to the exercise. Subject 44, also had short lower extremities but her exercise metabolic rate was plus 116 per cent.

Subject 11 was taken out of the normal group because he was a thin, physically inferior, individual with a kypho-scoliosis and a pigeon breast. The vital capacity of his lungs was also considerably diminished considering his surface area—76 per cent. It will be noted that the ratios of his basal and exercise oxygen consumption were, upon two occasions, 1.293 and 1.294 respectively. His recovery period was normal—three minutes—probably because his circulation was adequate.

Subject 14, apparently was normal in every respect except that his metabolic rate was consistently and abnormally diminished. His

basal-exercise oxygen ratio, however, was higher than the average—1.286. Subject 9, could not be satisfactorily diagnosed, but he was considered a case of psychoneurosis. It is of interest that, although he consumed relatively little oxygen during exercise he likewise required a correspondingly small amount during rest, and had a basal-exercise ratio of 1.192 which is less than the average for normal individuals. One case of myxoedema, which was of five years standing, showed a ratio of 1.28.

This exercise did not produce any striking increase in the pulse rate with the exception that, as would be expected, the cardiac and hyperthyroidism patients consistently showed an increase of about fifteen beats. The majority of the other subjects did not show an increase in the pulse rate of over six beats, while a few increased up to twelve. No observations were made on the length of time required for the pulse rate to return to its basal rate. Records of the vital capacities on all subjects have also been recorded, but so far as we could determine, no consistent correlation could be made.

There are included in the miscellaneous group, seven cases of heart disease, mostly with valvular lesions, who had but slight heart failure without marked decrease in the vital capacity of the lungs, with the exception of subjects 6 and 30. In four of these cases (5, 19, 37, and 1) there appeared to be an increase in the oxygen consumption during exercise considerably above what we had found in the other groups. It may be significant that the recovery periods of these four were found to be longer than we observed in other patients, which probably was a manifestation of their inadequate circulation. This corresponds somewhat to the observations of Peabody et al. who found that the return to normal of the minute-volume of the respiration and heart rate were delayed in cardiac patients following exercise as compared to normal individuals. The recovery to a normal resting oxygen consumption is probably dependent upon the same phenomena as the return to a normal pulmonary ventilation. Similarly we observed the same slight delay in the recovery period in two cases of mild hyperthyroidism.

SUMMARY

The individuals composing all the groups showed a rather wide variation in their response to exercise as far as oxygen consumption

was concerned. These are probably dependent upon factors, which, without present methods of study, are beyond our reach, and possibly are related only to the individual. Yet, despite this variation the averages of the exercise metabolism in two groups—those who were thin or of normal weight and those who were obese or overweight—agreed very closely, 121.9 and 124.4 per cent above the basal rate, respectively. This would indicate that it costs the overweight individual absolutely more and relatively just as much energy to move his lower extremities as it does the individual who is thin, or of normal weight.

As far as our observations could lead us, it appears that the basal metabolic rate was the most influential single factor in determining the amount of oxygen used during the exercise. The evidence points to the probability that each person has an individual rate of oxygen consumption during exercise and that his basal-exercise metabolism ratio is constant even though his basal metabolic rate should change.

The observations upon the length of time required for the subjects to pay off their oxygen debt after exercise suggest that such observations might be of value in the study of cardiac patients, in which group we found the recovery period to be quite uniformly prolonged.

While there was close agreement in the percentage increase of the exercise metabolism in the various groups, there was usually such a wide individual variation that the study has not yielded as much information with regard to a standard response as was hoped. Possibly with refinements of study and closer analysis of the subjects, from all points of view, the test might be helpful.

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EFFECT OF PHENYLHYDRAZINE DERIVATIVES IN THE TREATMENT OF POLYCYTHEMIA

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(Received for publication January 22, 1926)

The use of phenylhydrazine derivatives as therapeutic agents was introduced soon after the discovery of acetyl phenylhydrazine (pyro-din) by Liebreich. It was employed as an antipyretic. Renvers (1), in discussing this drug, pointed out that while its action as an antipyretic was fairly efficient, its toxic action upon the red blood cells made its use dangerous. He describes a case of rheumatic fever in which pyro-din grams 0.5 was given for three days, following which the red blood cell count dropped to two million eight hundred thousand cells. A distinct icterus developed and at the same time the urine became burgundy colored and contained much urobilin. A series of similar ill effects was noted by other observers who used the phenylhydrazine derivatives as antipyretics. For many years these drugs were employed only in the production of experimental anemias in animals and it was not until 1908 that Morawitz and Pratt (2) suggested that phenylhydrazine hydrochloride should be given a trial in polycythemia.

Sometime later Eppinger and Kloss (3) reported that they had obtained satisfactory results with this drug, using at first 10 cc. of a 5 per cent solution injected subcutaneously in a case of polycythemia. Further observation convinced them that oral administration was as effective as subcutaneous injection. A reduction of the red blood cells and hemoglobin with an occasional leukocytosis was noted.

Taschenberg (4) also used the drug in polycythemia with a satisfactory decrease of the red blood cells and a fall in the percent of hemoglobin to normal limits. His observations led him to believe that a specific resistance to phenylhydrazine was developed by the red cells which necessitated the administration of increasingly large doses to bring about the desired results.

The untoward effects of the therapy were pointed out by Strassburger (5) who treated two cases of polycythemia with phenylhydrazine hydrochloride. In his first case, as a result of too intensive treatment, the red count dropped to 2,600,000, with a hemoglobin of 64 per cent, and the patient felt very weak at this period. His second case, treated with smaller amounts was greatly benefited by the therapy. Geisbock (6) described a patient suffering from polycythemia hypertonica with a systolic blood pressure of 180 mm of mercury. Under treatment with phenylhydrazine hydrochloride the patient's red blood count was reduced to normal and the systolic pressure lowered to 120 mm.

At the Johns Hopkins Clinic, Owen (7) treated one case with excellent results. The red blood cells and hemoglobin reached a point somewhat below normal, with a simultaneous relief of the subjective symptoms. After three months, however, there was a recurrence of symptoms and the patient returned for further treatment. The second course of treatment showed that with the original dosage the red blood count could again be controlled as in the first course of treatment. Levi (8) believed that cirrhosis of the liver is accentuated by blood destruction and he advised against the use of phenylhydrazine in patients suffering from polycythemia.

In the Thorndike Memorial Laboratory, phenylhydrazine derivatives have been administered to three patients with polycythemia and one patient with hypertension. The observations here reported are the results of a study of the therapeutic dosage, the mode of action of the drug and the benefits derived therefrom. It is recognized that the series is not sufficiently large to warrant general conclusions, but certain actions of the drug seem very constant and are illustrated in the case reports that follow.

Case I F S A Jewish housewife, aged 68, entered the hospital August 24, complaining of pains in the arms, legs and chest. Her past and family history were unimportant. The skin of the patient's face and hands had been a dusky red color for eighteen years. In 1918 she began to have pains in her long bones and at that time a diagnosis of polycythemia vera was made. She had x-ray and radium therapy over her long bones with no improvement in her condition. The pain had steadily increased until the time of entry into the hospital.

The physical examination showed a poorly nourished and poorly developed woman who was complaining bitterly. The skin of the entire body, and especially

that of the hands and feet, had a dusky red tint. Numerous spider angiomas occurred in the skin of the face. The conjunctivae were moderately injected. The mucous membranes of the mouth were a slaty blue color. Her tongue was not atrophic. The heart and lungs were normal. A moderate smooth peripheral arterio-sclerosis was present. Blood pressure—130/70. The spleen was not felt.

TABLE 1

Date	Hemo- globin	Red b'ood cells	White b'ood cells	Reticulated cells	Plasma bilirubin	Drug
	<i>per cent</i>	<i>millions</i>	<i>thousands</i>	<i>per cent</i>	<i>mgm</i>	<i>grams</i>
August 19	135	10 0	5 0		0 4	
August 21	(Venesection 500 cc.)				0 44	
August 25	139	8 3		0 2		
August 27	(Venesection 500 cc.)					
August 27	133	8 4				
September 2	135	8 5				
September 10	133	8 2				
September 16	125	7 3	4 2	0 3	0 44	
September 16						0 1
September 17	126	7 3	5 6			0 1
September 18	131	7 3	6 7			0 1
September 19	121	6 6	6 5			0 1
September 20	115	6 4	5 6			0 1
September 21	112	6 6	5 6			
September 22	112	6 5	6 8	1 3	1 0	0 1
September 23	108	6 4	7 9			0 1
September 24	105	6 0	9 6			0 1
September 25	99	5 9	6 1			0 1
September 26	92	5 4	6 8			
September 27	93	5 6	6 9			
September 28						0 1
September 28	93	6 0	8 0			0 1
September 29	91	4 6	7 4		0 8	0 1
September 30	90	5 3	6 0	2 1		
October 1	82	4 8	10 3			
October 2	72	5 4	8 6			
October 3	76	5 5	8 8			
October 4	74	4 0	6 8			
October 5	71	4 2	6 9			
October 6	76	4 6	6 1	3 7	0 9	

Laboratory data Red blood cells—8,360,000 Hemoglobin (Newcomer-Haldane)—139 per cent White blood cells—5,000 The smear showed slight achromia Reticulated red blood cells—0.2 per cent Hematocrit—62 per cent Blood bilirubin (Van den Bergh)—0.4 mgm per 100 cc of plasma with a delayed direct diazo reaction The blood Wassermann was negative. The gastric

contents showed free hydrochloric acid The urine and stool were normal Basal metabolism (Aub-Dubois)—+12 per cent Diagnosis Polycythemia vera

As a preliminary measure the patient received two venesections during which a liter of blood was removed, but the red blood cell count and hemoglobin were not affected Three weeks after the last venesection, treatment with phenylhydrazine hydrochloride in doses of 0.1 gram per day was started

As will be noted in table 1, there was a rapid decrease in the hemoglobin content of the blood and in the total number of red blood cells The changes in the subjective symptoms of the patient did not parallel the blood picture, and it was not until several days after the drug was discontinued that the pains in the long bones and thorax disappeared The urine, which before the administration of the drug had been a light straw color, became a dark amber color soon after the treatment was instituted A moderate reduction of Benedict's Solution was noted on the first day and urobilin made its appearance on the third day There was no albuminuria or hematuria at any time

The patient was discharged greatly improved with instructions to return for further observations in a month's time Nothing more was heard of her until May, 1925, when she entered the hospital complaining of pain in the long bones and epigastrium After leaving the hospital in October, 1924, she remained in good health for three months, when a recurrence of symptoms became evident She obtained phenylhydrazine hydrochloride capsules from the outpatient department of another hospital, and instead of following the instructions given there took seven capsules in as many days Very little relief was obtained, however, and the pain so steadily increased that she decided to re-enter this hospital for treatment

At the time of the second entry the physical examination had not changed The red blood cells numbered ten million Hemoglobin (Newcomer-Haldane)—130 per cent White blood cells—9,800 The smear showed a slight achromia Reticulated red blood cells—0.1 per cent Hematocrit—58.5 per cent Blood bilirubin (Van den Bergh)—0.5 mgm per 100 cc of plasma with a delayed direct diazo reaction The blood Wassermann was negative The gastric contents showed free hydrochloric acid The urine and stool were normal

TABLE 2

Date	Hemo- globin	Red blood cells	White blood cells	Reticulated red cells	Plasma bilirubin	Drug
	<i>per cent</i>	<i>millions</i>	<i>thousands</i>	<i>per cent</i>	<i>m_μm</i>	<i>grams</i>
May 18	130	9 3	7 8	0 2		
May 20	140	8 8	12 1			
May 21	139	9 4	12 1			
May 22				0 8	0 5	0 1
May 23	140	8 9	9 4			0 1
May 24	145	9 6	7 9			0 1
May 25	135	11 1	10 8			0 1
May 26	125	8 4	11 9	19 0	0 6	0 1
May 27	125	8 4	10 6			0 1
May 28	135	7 4	7 5			0 2
May 29	130	9 8	11 2			0 2
June 1	125	7 9	11 5			0 2
June 2	125	7 4	11 9			0 2
June 3	109	6 4	9 6	39 0	1 4	0 2
June 4	109	6 4	9 0			0 2
June 6	101	6 5	12 2			
June 7	98	5 6	18 0			
June 8	93	5 9	15 6			
June 10	62	3 8	16 1		3 6	
June 11	60	3 0	24 0			
June 13	55	2 9	19 6			
June 14	52	2 4	17 0			
June 17	24 5	1 8	19 8			
June 18	25 0	1 7	20 2	34 0	1 2	
June 19	29	1 5	22 0	46 0		
June 21	31	1 9	18 4	43 0		
June 22	31 0	1 7	14 2		4 2	
June 23	28 0	1 5	9 8	40 0		
June 25	28 4	1 4	7 7			
June 26	28	1 3	9 0			
June 26	(Transfused 500 cc. citrated blood)					
June 27	36	2 1	7 6			
June 30	41	2 4	7 0	10 0		
July 2					0 2	
July 4	48	2 3	4 4			
July 7	46	2 5	7 0	5 0		
July 12	46	2 3	8 5	1 0		
July 19	72	3 9	9 7			

Treatment with phenylhydrazine hydrochloride was instituted as is shown in table 2. As the patient seemed rather resistant to treatment, it was thought advisable to increase the drug gradually. The

phenylhydrazine was stopped when the hemoglobin reached 101 per cent. Despite the cessation of treatment the blood continued to fall rapidly. With the decrease in the red count the patient became very weak and she suffered constantly from gastric distress. Because of her prostration and gastric distress the decision was made to transfuse her, following which procedure an improvement was noted within a short time. Her strength returned and the nausea and vomiting ceased. The red blood cell count slowly increased until she was discharged, when the blood picture was approaching normal.

Case II G W A white American shoe inspector aged 63, entered the hospital September 21, 1924, complaining of a partial paralysis of the right side. His family and past history were unimportant. The patient stated that in 1921 his friends remarked about his "high color." He had felt very well until November, 1922, when he had a monoplegia of his left leg. He entered a hospital and remained there six weeks. He returned to work and remained in good health until May, 1923, when he had a right-sided hemiplegia. The patient entered a hospital and stayed there six months, during which period he suffered from severe headaches. He was told that he had "too much blood" and frequent venesections were performed with no relief. His hemiplegia improved and he was able to walk with the aid of crutches, but throughout the past year he has suffered from daily headaches.

The physical examination revealed a well nourished man, oriented in all respects. The skin of the face and hands had a beefy red hue. There were many small spider angiomas over the maxillary prominences. The conjunctivae and the mucous membranes of the mouth were injected. The tongue was not atrophic. The heart and lungs were normal. A moderate amount of smooth sclerosis was present in the radials, brachials, and temporals. Blood pressure—148/96. Residual findings of a right hemiplegia were present.

Laboratory data Red blood cells—10,230,000 Hemoglobin (Newcomer-Haldane)—122 per cent White blood cells—18,300 The smear showed slight achromia Reticulated red blood cells—0.8 per cent Hematocrit—62 per cent Blood bilirubin (Van den Bergh)—0.3 mgm per 100 cc of blood plasma The urine and stool were normal. Diagnosis Polycythemia vera.

Inasmuch as the red blood cell count of this patient did not diminish under the original dosage it was deemed advisable to increase the amount of phenylhydrazine as is shown in table 3. This was done without producing any untoward events. The clinical condition of the patient improved from the beginning of his treatment. His headaches soon disappeared and he gradually became stronger. A

TABLE 3

Date	Hemo- globin	Red blood cells	White blood cells	Reticulated red cells	Plasma bilirubin	Drug
	<i>per cent</i>	<i>millions</i>	<i>thousands</i>	<i>per cent</i>	<i>mg.</i>	<i>grams</i>
September 25	122	10 2	18 3	0 8	0 3	
September 29	128	9 8	14 7		10 4	0 1
September 30	133	10 0	17 5			0 1
October 1	124	9 7	17 4			0 1
October 2	118	9 6	15 8			0 1
October 3	118	9 8	16 8			0 1
October 4	120	9 8	18 0			0 1
October 5	118	10 0	17 1			0 1
October 6	116	9 4	15 1	1 4	0 8	0 1
October 7	125	10 0	12 4			0 2
October 8	123	8 7	18 1			0 1
October 9	125	9 1	16 4			0 2
October 10	135	9 8	17 9			0 1
October 11	133	10 2	17 4			0 2
October 12	125	9 9	18 6			0 1
October 13	127	8 2	16 1		0 7	0 2
October 14	125	8 7	15 3			0 1
October 15	125	8 9	26 2			0 2
October 16	125	8 4	21 3			0 2
October 17	127	9 0	20 8			0 2
October 18	116	9 4	21 8	1 4		0 2
October 20	114	8 8	14 9		0 9	0 2
October 21	122	9 7	20 4			0 2
October 22	122	9 0	16 4			0 2
October 23	122	9 1				
October 24	116	9 3	16 8			
October 25	116	7 9	20 4		1 2	
October 27	111	7 9	17 6		1 2	
October 28	114	8 6	24 4			
October 29	108	8 4	25 0			
October 30	111	8 3	22 0			0 4
October 31	108	8 2	23 0			0 4
November 3	106	7 6	26 1	3 4	1 4	
November 4	106	7 3	24 1			
November 5	102	7 1	20 6			
November 6	106	7 0	21 9			
November 7	104	6 5	22 3			
November 8	102	6 0	26 9			
November 10	101	5 9	24 0		1 1	
November 11	97	5 9	24 5			

phlebitis of the internal saphenous veins of both legs was the only incident during the whole period of treatment and thus cleared up rapidly under symptomatic therapy

The urine which was straw-colored prior to the administration of phenylhydrazine, soon developed a dark amber color and caused a moderate reduction of Benedict's Solution throughout the period of treatment. Urobilinogen was present from the third day until

TABLE 4

Date	Hemo- globin	Red blood cells	White blood cells	Reticu- lated red cells	Plasma bilirubin	Drug	Blood pressure
	<i>per cent</i>	<i>millions</i>	<i>thou sands</i>	<i>per cent</i>	<i>mgm</i>	<i>grams</i>	<i>mm Hg</i>
December 30	92	5 2	8 7			0 1	260/140
December 31	92	5 4	8 6			0 1	
January 2	89	5 3	8 2			0 2	240/130
January 3	84	4 8	8 5			0 1	
January 4	84	5 1	8 3			0 1	240/150
January 7	81	4 4				0 1	
January 9	80	4 2				0 2	220/130
January 13	78	4 0				0 1	
January 15	61	3 3					180/90
January 16	58	2 3					
January 17	42	2 3					
January 18	34	2 0	8 9	13 6			
January 20	34	2 0	9 6	14 4	1 2		180/90
January 22	34	2 3	7 8	17 3			
January 24	41	2 7	7 9	13 2	0 9		190/80
January 26	43	2 7	6 3	15 8			
January 29	51	2 5	4 4	10 4			140/80
February 1	70	3 4					
February 3	61	3 1		4 3			
February 8	80	3 7		1 8			195/110

three weeks after the cessation of the drug. There was no albuminuria or hematuria at any time. Methemoglobin was found in the blood spectroscopically during treatment. This patient was discharged completely relieved of all symptoms in any way referable to his polycythemia.

The beneficial results obtained by Geisbock in a case of polycythemia hypertonica treated with phenylhydrazine hydrochloride led us to test the efficacy of the drug in a case of Essential Hypertension

Case III M S A white American housewife, aged 60, entered the hospital December 9, 1924, complaining of weakness in the left leg. Her mother and maternal grandmother died of apoplexy. For fifteen years previous to her entry she had been troubled with indigestion. One year before coming to the hospital a cholecystectomy was done. Four years before she had been told that she had "high blood pressure." She never had headaches. For a few months before entrance she had been troubled with nocturia and frequency. On December 4, 1924, her left leg became weak while walking, and on arising the next morning she found it impossible to use that leg. At that time she suffered from numbness in her left hand and arm. This weakness and numbness had practically disappeared before coming to the hospital.

The physical examination showed a well developed and well nourished woman. Except for the cardio-vascular system no important abnormalities were found in the examination. The heart was slightly enlarged, but the sounds were regular and of good quality. There were systolic murmurs at the apex and in the aortic area. The pulses were regular and equal. The radial and brachial arteries were moderately sclerosed and were easily palpable. Blood pressure—270/160. No definite residual signs of the hemiplegia could be detected.

Laboratory data Red blood cells—5,250,000 Hemoglobin (Sahli)—92 per cent. White blood cells—7,600. The smear was normal. Blood bilirubin (Van den Bergh)—normal. The blood Wassermann was negative. The urine and stool were normal. Phenolsulphophthalein—45 per cent, in two hours. Diagnoses: Arteriosclerosis, hypertension.

As is shown in table 4, there was a marked change in the patient's blood pressure while under phenylhydrazine therapy. While there was a drop in both the systolic and diastolic pressures, it was believed that this might be attributed to the fact that the patient was very quiet during the period when the red blood count was low. It was interesting to note that during the period of anemia the patient insisted that she felt better than she had for months.

The urine was a dark amber color and contained urobilin. There was no albuminuria or hematuria at any time. Methemoglobin was determined in the patient's blood one week after phenylhydrazine was started. There were no irritating symptoms attending the use of the drug. A normal regeneration of the blood accompanied by the pouring out of reticulated cells and normoblasts took place. At times, myelocytes were present. When discharged the patient's blood had gradually returned to normal.

Case IV P M A Russian laborer, aged 36, entered the hospital February 27, 1925, complaining of shortness of breath, weakness and headaches. His past

and family history were unimportant. Three years before his entrance he noticed that he tired easily and a short time later nocturnal headaches began to occur. His friends remarked upon the redness of his face and hands. He stopped work but the weakness and headaches became more pronounced, and he soon developed difficulty in getting his breath. He entered a hospital and from there was sent to a convalescent home for several months. His rest benefited him but as soon as he started to work, he again became very weak and short of breath. Once more he entered a hospital and was discharged in a few weeks unimproved. Shortly after this he came under our observation.

The physical examination showed a well developed and well nourished man lying in bed without any apparent discomfort. The skin of his face and hands was a dusky blue in color. His conjunctivae were injected and the mucous membranes of his mouth and lips were very cyanotic. There was no atrophy of the mucous membranes of the tongue. The thorax showed marked enlargement in the antero-posterior diameter and was typically barrel-shaped. His thorax moved as a whole during respiration and the respirations were slow and regular. There was no dyspnea. The lungs were hyperresonant to percussion. Tactile and vocal fremitus were normal. The breath sounds were emphysematous in type. The lower border of the lung descended 1.5 cm. on full inspiration on the right side, and 2 cm. on the left. The maximum impulse of the heart could be neither seen nor felt, and the area of cardiac dullness could not be made out. The sounds were distant, low and regular, and there were no murmurs. The pulses were equal, rhythmical, and of good tension. There was no peripheral arteriosclerosis. Blood pressure—122/84. The abdomen was normal. There was a suggestion of clubbing in the distal phalanges of the fingers. Fluoroscopy showed that the thorax moved little, if at all, during inspiration. Roentgenograms revealed that the heart was enlarged in the region of the left auricle and that the pulmonary artery was enlarged. The lungs showed emphysema in the lower lobes and few bronchial markings in the upper lobes. The electrocardiogram showed normal rhythm, P R—0.20 second, P₂—0.1 millivolt, Q R S—0.08 second. Axis normal. Variation in S₂ and T₂. The vital capacity was, sitting—3,100 cc., standing—3,000 cc., lying—2,550 cc.

Laboratory data Red blood cell count—6,240,000. Hemoglobin (Newcomer-Haldane)—120 per cent. White blood cells—7,400. The smear was normal. Reticulated red blood cells—0.2 per cent. Hematocrit 53 per cent. Blood bilirubin (Van den Bergh)—0.4 mgm. per 100 cc. of blood plasma with a delayed direct diazo reaction. The Wassermann was negative. The sputum showed no tubercle bacilli. The urine and stool were normal. The phenolsulphophthalein test returned 50 per cent of the dye in 2 hours and 10 minutes. Basal metabolism (Aub-Dubois)—plus 3 per cent. Diagnosis Emphysema with compensatory polycythemia or Ayerza's syndrome.

With this patient, it was a question as to whether the polycythemia was the primary condition. It was our desire to observe whether

any beneficial effects would be obtained if the patient's blood picture was reduced to normal. Acetyl phenylhydrazine (pyrodim) was used because Bodansky (9) reported that it seemed to be the least toxic member of the phenylhydrazine series.

Table 5 shows that the hemoglobin and red count gradually declined under the influence of the drug. Daily differential white blood

TABLE 5

Date	Hemo- globin	Red blood cells	White blood cells	Reticulated red cells	Plasma bilirubin	Drugs (pyrodim)
	<i>per cent</i>	<i>millions</i>	<i>thousands</i>	<i>per cent</i>	<i>mgm</i>	<i>grams</i>
April 6	120	7.4	7.8	0.2	0.35	0.1
April 7	113	7.3	9.3	0.1		
April 8	116	6.8	6.3		0.33	0.1
April 9	104	6.2	6.0			0.1
April 10	102	6.4	10.1			0.1
April 11	92	6.2	7.1			0.1
April 13	84	5.2	7.2	0.2	0.5	0.2
April 14	85	6.2	9.5			
April 15	90	5.9	5.5			0.1
April 16	93	5.3	6.5			0.1
April 17	81	5.6	5.2			0.2
April 18	84	5.2	4.8			0.2
April 19	94	5.7	8.7			0.2
April 20	81	5.2	6.2			
April 21		5.1	8.3	1.3		0.2
April 22	80	4.5	5.5			
April 23	69	4.5	7.1		0.9	
April 25	75	4.6	5.7			
April 27	72	4.3	8.5			
April 28	78	4.5	9.5	6.8		
April 29	74	4.2	7.4		1.1	
April 30	78	4.5	9.0			
May 1	79	4.2	9.1			
May 2	83	4.5	9.0			
May 4	89	4.0	9.0			
May 6	93	5.7	7.8	4.4	0.9	
May 13	85	4.5	7.4	0.8	0.7	

counts were made using the vital technique as described by Sabin (10). It is interesting to note that on three successive days numerous monocytes with ingested red blood cells were observed. On one occasion a small clasmatocyte containing hemoglobiniferous particles was seen in the peripheral circulation.

The urine partially reduced Benedict's Solution on the first day of the therapy and continued to do so until eleven days after the drug had been stopped. Urobilin appeared in the patient's urine upon the fourth day and was present in large amounts until the time of patient's discharge from the hospital. There was no hematuria or albuminuria at any time.

For the first few days the patient seemed to be benefited by the decrease in the hemoglobin content of the blood. He said that he felt stronger and that his headaches and shortness of breath were markedly relieved. This improvement continued until the hemoglobin dropped below 80 per cent, at which point the patient began to complain of great weakness, shortness of breath, nausea and vomiting, and headache. The drug had been discontinued two days before these distressing symptoms put in their appearance. Ten days later when the blood count and hemoglobin had increased, the patient made some improvement, but not enough to warrant the use of phenylhydrazine in this type of polycythemia.

One must be guided by the needs of the patient when using phenylhydrazine derivatives in the treatment of polycythemia. Experience in this hospital indicates the wisdom of using small doses of the drug in the beginning of the treatment. It seems wise to start with 0.1 gram per day by mouth and to continue this amount until the red blood count begins to fall or until one gram of the drug has been given. Then it seems well to discontinue the drug for three days and observe whether or not its cumulative action is marked. If there is no significant change in the blood picture the drug should be given again, alternating 0.1 gram and 0.2 gram doses until the desired results have been obtained or until another gram of phenylhydrazine has been administered. A week should pass before starting the third course of the drug if two grams do not bring about the desired results. The red blood cell count, hemoglobin and white blood cell count should be determined daily in order that any sudden change in the blood picture may be detected at once. By proceeding slowly it is possible to obtain the maximum therapeutic effect of phenylhydrazine with little danger to the patient.

Phenylhydrazine hydrochloride, when taken by mouth in capsules, did not cause irritation of the gastro-intestinal tract. It was ab-

sorbed into the blood and caused methemoglobin to be formed. This apparently injured the red blood cells and allowed them to be phagocytized and then destroyed. The white blood cells did not seem to be affected by the phenylhydrazine, as no leukocytosis referable to phenylhydrazine was noted. Any change in the white blood cells fell within the limits of the normal rhythm of the leukocytes (11). The differential white blood cell count remained normal under this treatment. The reticulated red blood cell count increased as the red blood cell count dropped. Occasionally normoblasts and myelocytes were seen if the blood dropped to a low level. The platelets remained normal. Phagocytosis of red blood cells in the peripheral blood stream occasionally was seen. Coincident with this phagocytosis the patients developed slight jaundice and the blood bilirubin was increased in amount and gave the delayed direct diazo reaction. The bilirubin content of the blood gradually declined after treatment was stopped, and after blood destruction ceased.

The color of the urine changed soon after the drug was administered, and assumed a deep amber color. The phenylhydrazines are reducing agents, and mild reducing substances have been noticed in the urine of the patients under treatment. Urobilinogen and urobilin appeared in the urine on the second or third day of treatment and continued to be present as long as blood destruction took place.

No evidence of kidney irritation has been observed in any of the patients who received phenylhydrazine hydrochloride. Apparently the drug had a moderate cumulative action, being slowly excreted in the urine. This cumulative action upon the blood was shown by the fact that the red blood cell count continued to fall for several days after the drug had been discontinued.

The best results with phenylhydrazine were achieved in the treatment of polycythemia vera. In this condition, with no discomfort to the patient, the blood can be reduced to normal and held there with ease for a considerable period of time. With a normal blood picture these patients lose the symptoms of their polycythemia and are greatly benefited. We have not seen any patient in whom we have detected any signs of liver damage arising from phenylhydrazine.

In our experience the drug was of little value in the treatment of secondary polycythemia or hypertension.

CONCLUSIONS

Phenylhydrazine hydrochloride and acetyl phenylhydrazine are efficient therapeutic agents in the treatment of polycythemia vera

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EXPERIMENTAL ANEMIA PRODUCED BY PHENYLHYDRAZINE DERIVATIVES

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(Received for publication January 22, 1926)

Soon after the discovery of acetyl phenylhydrazine (pyrodin) by Liebreich it was observed that this chemical possessed marked antipyretic effects, and it therefore came into widespread use in diseases accompanied by fever. Further experience indicated that in addition to reducing the temperature, the drug also produced an anemia that varied in degree according to the dosage. Experiments performed by Dreschfeld (1) on the effects of pyrodin in rabbits showed that an anemia accompanied by hemoglobinuria was produced, and that many phagocytic cells containing intact erythrocytes and hemoglobiniferous particles appeared in the spleen. He also noted that spectroscopic examination of the blood showed the presence of methemoglobin.

There have been many observations upon the type of anemia produced by phenylhydrazine since Dreschfeld's early report. Ziegler (2), Lafleur (3), Wertheimer (4), Fraenkel (5), Albertoni (6), Gatti (7), Ballistini (8), Morawitz and Pratt (9), Gilberti (10) and Foti (11) have carefully described the peripheral blood changes produced by phenylhydrazine. Lafleur (3), Albertoni (6), Gatti (7), Morawitz and Pratt (9) examined the various organs affected by this drug both macroscopically and by histological methods. Suzuki (12) and Taschenberg (13) reported increased resistance on the part of the red blood cells to solutions of phenylhydrazine. In 1915 Eppinger and Kloss (14) recommended the use of phenylhydrazine hydrochloride in the treatment of polycythemia vera. They found that under its influence they could reduce the number of red blood cells and the percentage of hemoglobin to normal, and with this reduction in cell count an amelioration of symptoms took place. These observations stimulated interest in the use of phenylhydrazine deriva-

tives, and in view of the fact that no complete morphological and physiological study has been made, it seemed that a careful investigation of the effect of these substances upon the blood and blood forming organs in animals might be of value

METHODS

Adult rabbits weighing about 2 kg were used, and daily studies of the blood were made upon each animal. These studies consisted of total white blood cell counts, total red blood cell counts, hemoglobin determinations (Newcomer) (15), hematocrit determinations, reticulated red blood cell counts, and differential counts after the vital technique of Sabin (16). The fragility of the red blood cells of certain animals was tested against solutions of sodium chloride and phenylhydrazine hydrochloride. Rosenthal's (17) phenoltetrachlorophthalein test was used in determining the liver function. The presence of methemoglobin was determined spectroscopically. The bile pigments in the blood plasma were studied by Van den Bergh's (18) bilirubin method. At death studies of the fresh bone marrow, spleen and lymph nodes were made with the supravital dyes, neutral red and Janus green, according to the technique devised by Doan, Cunningham and Sabin (19). Histological studies of sections of the bone marrow, spleen, liver, lungs and kidney were made on autopsy material from each animal.

EXPERIMENTAL

In the first series 9 rabbits were used and after a control period of three days each animal received an injection of 30 mg of phenylhydrazine hydrochloride into the peritoneal cavity. One rabbit was then killed on each successive day.

Within twenty-four hours after the drug had been administered an anemia had begun to develop, and this anemia ran essentially the same course in all the animals. It increased in severity until the fourth day, at which time the destruction of red blood cells was balanced by red blood cell production. Following this the red blood cell production increased over destruction and the red blood cell count and hemoglobin returned towards normal. The details of the peripheral blood studies are shown in figure 1, which illustrates the

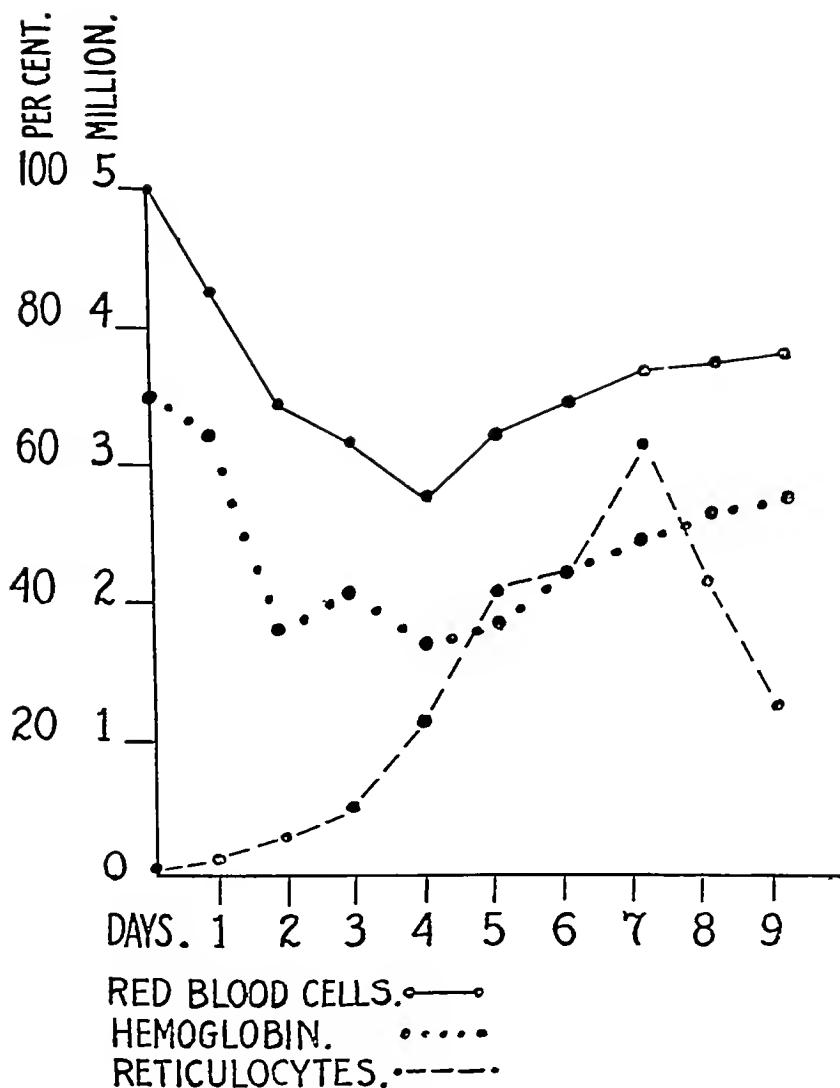


FIG 1

course of the anemia in a representative animal. At the period of most severe anemia the peripheral blood contained large reticulated red blood cells which numbered as high as sixty per cent of the

erythrocytes With the return of the hemoglobin and red cells to normal these reticulated cells decreased Very few microcytes or poikilocytes were seen in the peripheral blood and the color index remained constantly at about the normal level for rabbits Occasionally during the anemic stage monocytes containing hemoglobiniferous particles were observed in the peripheral blood

It has been suggested that the clinical administration of phenylhydrazine hydrochloride may be controlled by daily white bloodcell counts and that when a slight leukocytosis appears the drug should be stopped In experimental animals no definite leukocytosis was observed that could in any way be correlated with the injection of the drug

The resistance of the red blood cells to hypotonic solutions of sodium chloride and to phenylhydrazine hydrochloride was determined in 6 rabbits on the control days and on the third, sixth and ninth days of the anemia The resistance to hypotonic solutions of sodium chloride was determined according to standard technique In determining the resistance of the red blood cells to phenylhydrazine hydrochloride, 20 cu mm of blood taken from the marginal ear vein of the rabbit were placed in tubes containing 1 cc of freshly prepared phenylhydrazine hydrochloride solutions varying in strength from 0.20 to 0.68 per cent, and the tubes were immediately shaken to make a suspension The tubes were then allowed to stand for two hours at room temperature At the end of this time the tube showing a complete lack of corpuscular residue was recorded as the point of complete hemolysis It was found, as is shown in figure 2, that the resistance of the red blood cells to sodium chloride solutions remained the same throughout the anemia, but there was a marked decrease in the resistance of the red blood cells to phenylhydrazine on the third day of the anemia, followed by an increase in resistance on the ninth day—a marked increase over that of the control period

The liver function of each rabbit was determined by the phenoltetrachlorophthalein test of Rosenthal (17) just before the animal was killed In all the experiments the retention of the dye was approximately the same regardless of whether the animal had been injected with the drug one day or nine days before the test At the end of fifteen minutes about 3.3 per cent to 4.4 per cent of the dye remained

in the blood in contrast to 0.5 per cent in normal animals, but at the end of forty-five minutes the retention was only slightly above normal. Figure 3 shows curves of the average figures for each group of animals.

Methemoglobin was shown to be present in the blood by spectroscopic examination in 5 of the animals—on the third day of the

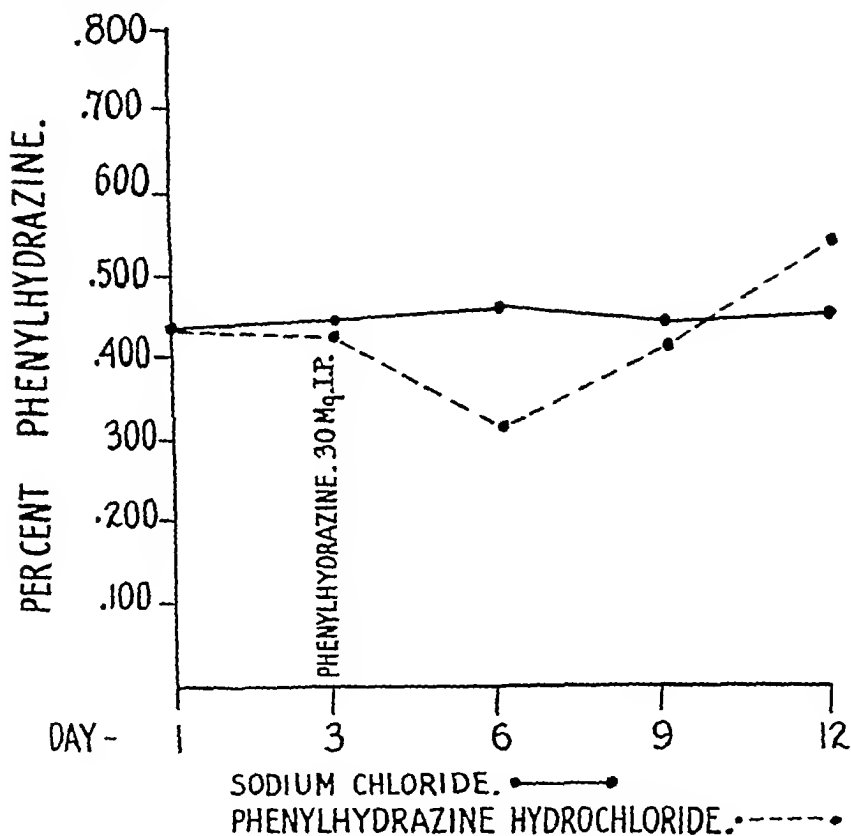


FIG 2

anemia. When a solution containing 440 mg of phenylhydrazine in 10 cc of Ringer's solution was injected into the ear vein of a rabbit, it was found that the animal died after the first 2 or 3 cc had been introduced. On examination the blood was found to be chocolate brown in color and gave a strong methemoglobin band in the spectrum.

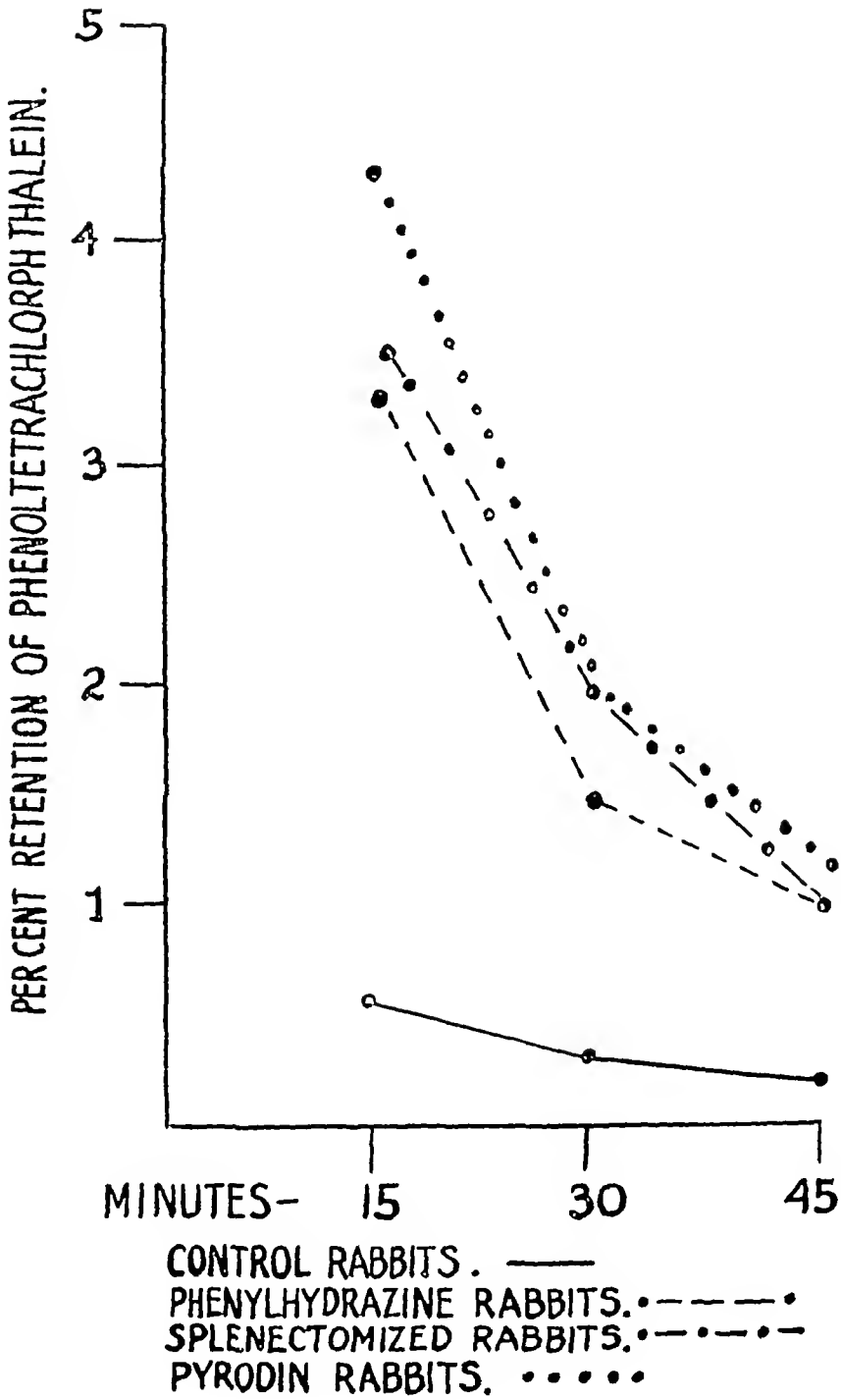


FIG 3

Blood bilirubin determinations were made on the blood plasma of several rabbits during the period of anemia, but no bilirubin was detected in the plasma. The experience of others, however, has shown that it is very difficult to demonstrate the presence of this pigment in the blood of rabbits. It is generally supposed that the threshold for bilirubin is low in these animals. A short time after the appearance of the anemia it was possible to demonstrate the presence of urobilinogen in the urine of the rabbits. The amount of urobilinogen increased until the eighth or ninth day of the anemia, after which there was a gradual decrease and it was not until three weeks after the injection of the phenylhydrazine hydrochloride that the urine was free from urobilinogen.

The animals were placed under ether anesthesia and just before death studies after the vital technique of Doan et al (19) were made upon fresh tissues from the bone marrow, spleen and mesenteric lymph nodes. The femoral bone marrow showed a hyperplasia of the red cell elements as the anemia progressed. There was a definite increase in the number of erythroblasts and normoblasts which on the seventh or eighth day of the anemia numbered as high as 80 per cent of the total cells. With the progression of the anemia the number of the earliest form of red blood cells—the megaloblast—increased until in the very hyperplastic marrows, on the six and seventh day, the megaloblasts formed 3 or 4 per cent of the total cells. In contrast to these observations, it was found in the femoral marrows of normal rabbit that no megaloblasts were seen, and that only 15 to 30 per cent of the total cells were erythroblasts and normoblasts. No hyperplasia of the myelocytic series was present in these animals. The clasmotocytes of the bone marrow increased in numbers reaching a total of 5 or 6 per cent, in contrast to 1 per cent in the normal controls. In the early stages of the anemia these cells were loaded with phagocytized red blood cells, but as the anemia progressed they contained mainly debris. The megakaryocytes were increased. It was interesting to note that the polymorphonuclear cells would stream up to the periphery of the megakaryocytes and slowly invade the outer rim of the cytoplasm of the giant cells. Once this rim was passed they would stream faster often going around and around the nucleus of the megakaryocyte. The polymorphonuclear leuko-

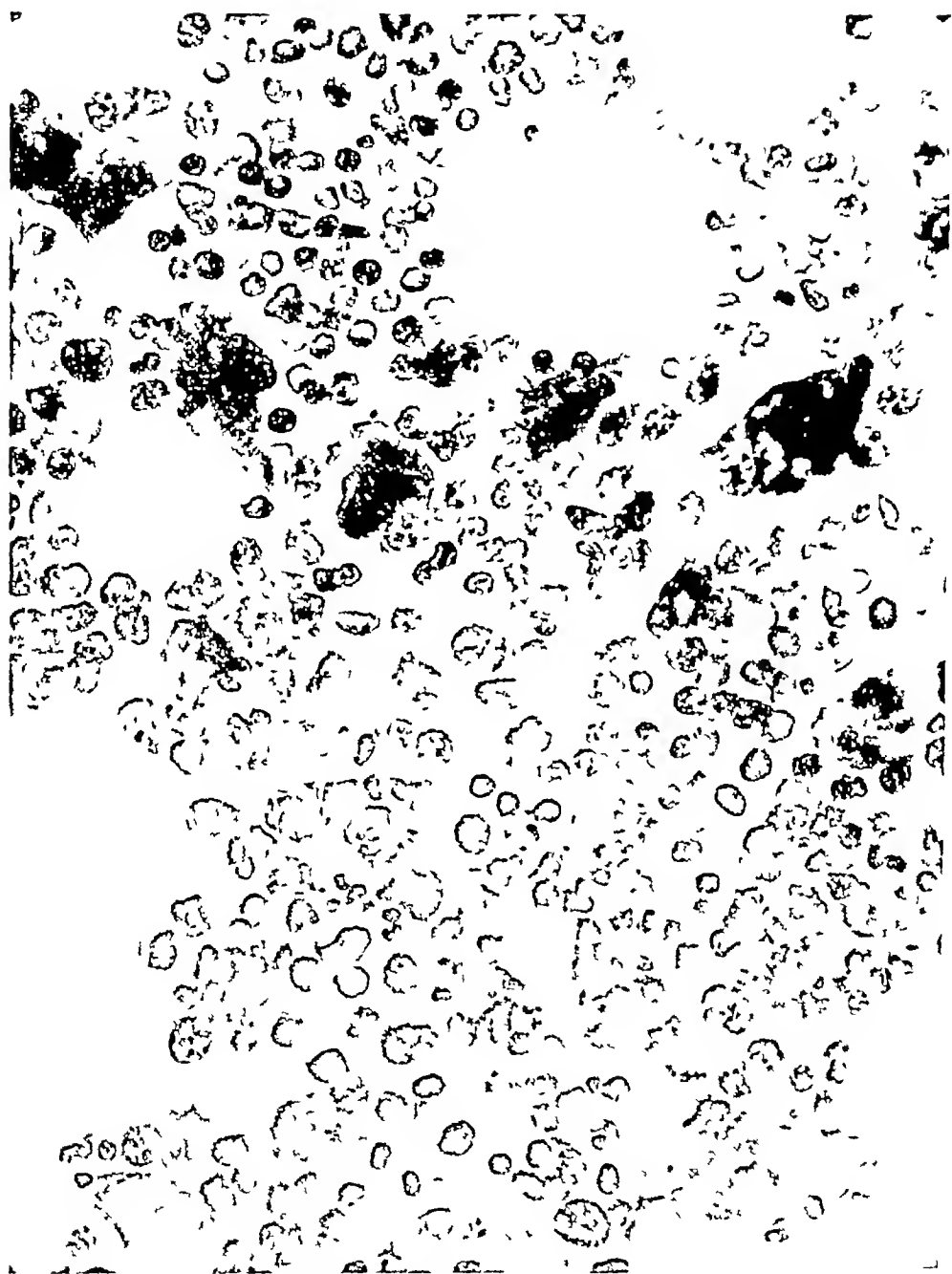


FIG 4 RABBIT VI POTASSIUM FERROCIANIDE AND FUCHSIN SECTION OF HYPERPLASTIC FEMORAL BONE MARROW SHOWING LARGE PIGMENT-BEARING CLASMATOCYTES WHICH GIVE THE IRON REACTION $\times 1000$

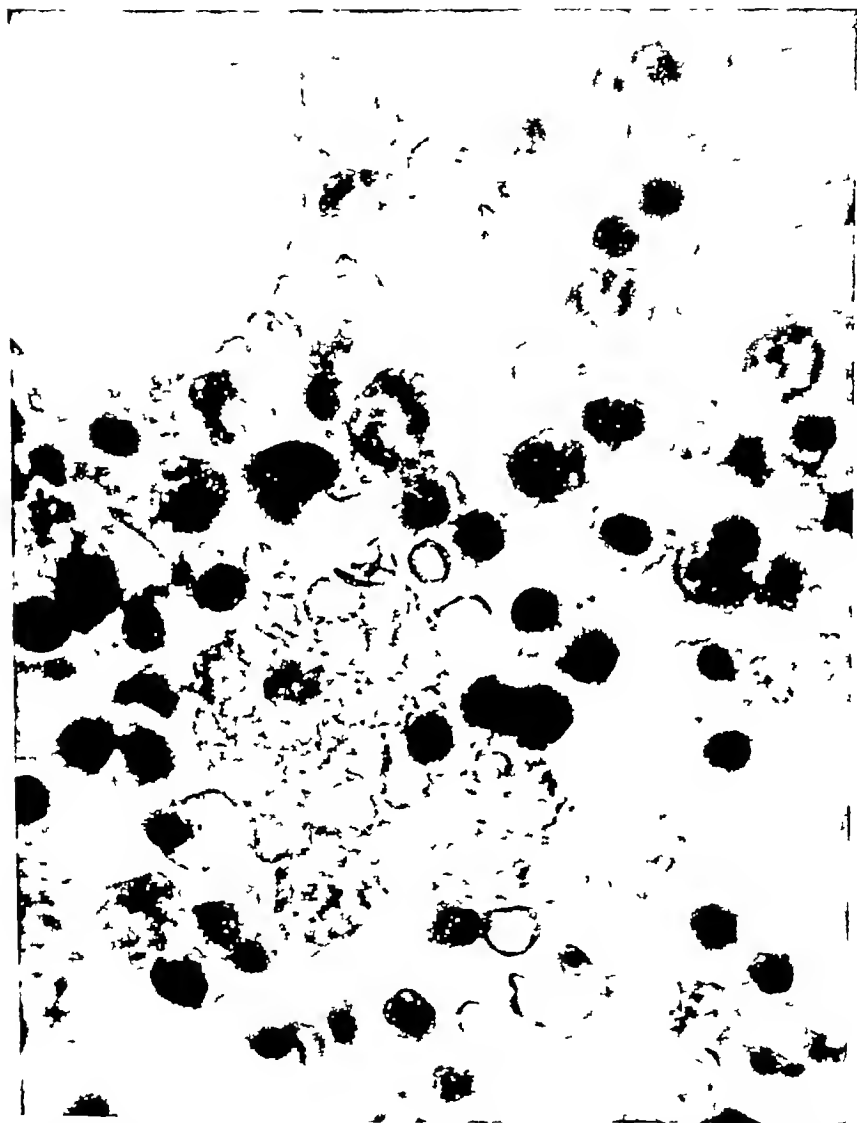


FIG 5 RABBIT VI LOSIN AND METHYLENE BLUE SECTION OF HYPE-
PLASTIC FEMORAL BONE MARROW SHOWING A LARGE CLASMATOCYTE PACKED
WITH RFD BLOOD CELLS / 2000

cytes, then, either became inactive in the cytoplasm of the giant cells or streamed off through the cytoplasm and passed out of the cell

The histological sections of the bone marrow also showed an increase in the erythrocytic series that became more marked as the anemia progressed. With the hyperplasia of the red blood cell elements there was a diminution of the fat cells, and on the eighth and ninth days of the anemia the marrow was practically a solid erythropoietic tissue. At all times, however, the blood sinuses of the bone marrow were wide open and packed with red blood cells. There was a marked increase in the clasmatocytes of the marrow, many containing red blood cells, all of the phagocytic cells also contained pigment giving an iron reaction with potassium ferrocyanide. The megakaryocytes increased in number with the hyperplasia of the erythrocytic tissue and often contained polymorphonuclear leukocytes. There were no constant changes in the leukocytic series.

The spleens were slightly enlarged but appeared otherwise normal. The studies of the living tissue showed a marked increase in the number of clasmatocytes. These increased rapidly as the anemia progressed and on the third or fourth day from 8 to 13 per cent of the cells from the spleen were clasmatocytes in contrast to the normal splenic count of 3 or 4 per cent. These cells were all packed with red blood cells, and some of them had as many as ninety red blood cells included in their cytoplasm. As the anemia progressed the red blood cells were broken down and on the eighth or ninth day the clasmatocytes contained debris. In the histological sections there were no constant changes in the lymphoid or fibrous elements of the spleen. There was, however, a marked increase in the number of clasmatocytes containing intact red blood cells, and a pigment which took the stain for iron.

The mesenteric lymph nodes gradually increased in size during the anemia until on the ninth day they reached three or four times their normal size. Supra-vital studies showed an increase in the reticular cells and in the lymphoblasts. In these nodes were many clasmatocytes which contained debris, but occasionally one would find phagocytic cells packed with erythrocytes. In the histological

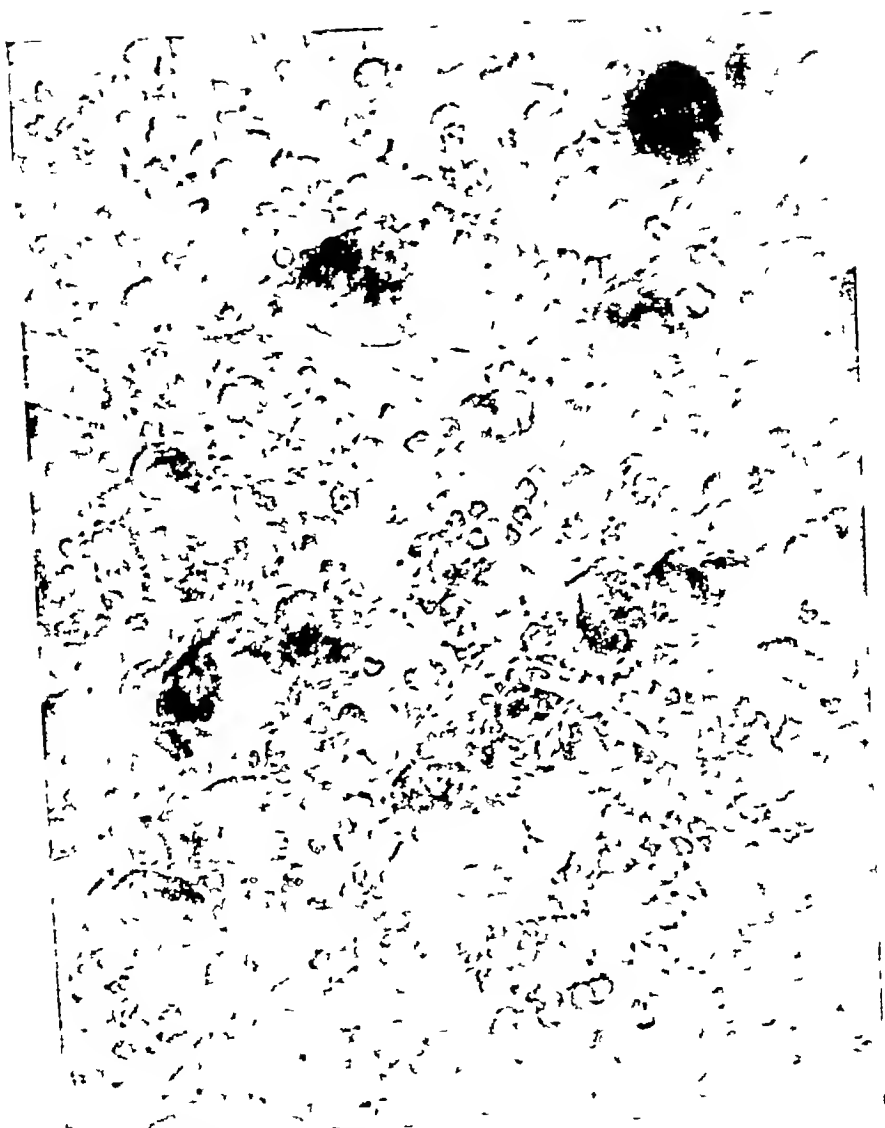


FIG 6 RABBIT XVII POTASSIUM FERROCYANIDE AND FUCHSIN SECTION
OF SPLEEN SHOWING NUMEROUS PIGMENT-CONTAINING CLASMATOCYTES
WHICH GIVE THE IRON REACTION $\times 1000$

sections there was a marked hyperplasia of the lymphoid tissue with numerous phagocytic cells, a few of which contained red blood corpuscles. The majority of the phagocytic cells contained a yellow pigment which did not give the iron reaction.

The histological findings in the livers were inconstant, but the liver parenchyma was apparently not damaged in any instance. There was no evidence of fatty change or necrosis. As the anemia progressed there was a gradual increase in the Kupfer cells, these cells containing particles which gave the iron reaction. The glycogen content of the liver as evidenced by Best's carmine stain was variable. There were no changes of any note in the lungs and kidneys.

In the second series of nine rabbits, acetylphenylhydrazine (pyrodim) was injected and the same technique as in the first series was followed. The changes in the peripheral blood were identical with those produced by phenylhydrazine hydrochloride. The liver function was determined and gave a slight retention of the dye at the end of fifteen minutes which fell to normal after forty-five minutes. Studies of living tissue and histological studies gave the same results as in the animals receiving phenylhydrazine hydrochloride.

The third series of experiments was done on animals after splenectomy. Phenylhydrazine hydrochloride was injected in the same manner as in the first series. The anemia produced was in no way different from that produced in normal rabbits. There was no change in the liver function curve. Studies of the living tissue and histological studies showed that with the extirpation of the spleen, the clasmatoocytes of the bone marrow and the Kupfer cells of the liver were increased in number and activity, thereby assuming the phagocytic function of the spleen.

SUMMARY

An anemia, pursuing a similar course in all the experiments, was produced by the intraperitoneal injection of phenylhydrazine derivatives in rabbits. When the anemia was established it was possible to demonstrate the presence of methemoglobin in the blood serum. There was a marked increase in the percentage and total number of reticulated red blood cells in the peripheral blood during the period

of the anemia, and the hematopoietic system underwent a marked hyperplasia which progressed until red blood cell production overbalanced red blood cell destruction. A marked increase in the number of phagocytic cells containing intact red blood cells and pigment was found in the spleen, bone marrow and lymph nodes during the anemia. It was not possible to demonstrate the presence of bilirubin in the blood plasma of the rabbits during the anemic stage. Urobilinogen appeared in the urine on the first day after the injection of phenylhydrazine and remained in demonstrable amounts for about three weeks.

After the administration of phenylhydrazine there was a decreased resistance of the red blood cells to solutions of phenylhydrazine which lasted until there was an outpouring of new red blood cells from the bone marrow. These young red blood cells showed an increased resistance to phenylhydrazine.

There was a disturbance in liver function as evidenced by the early retention of phenoltetrachlorophthalein but it did not seem to be marked. No histological changes denoting liver injury could be found in any of the rabbits.

There was no essential difference between the type of anemia produced by phenylhydrazine hydrochloride and acetylphenylhydrazine. The anemia produced in splenectomized animals did not vary from that produced in normal animals except that in animals with the spleens removed there was more phagocytosis in the bone marrow and liver.

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STUDIES ON HUMAN CAPILLARIES

I AN APPARATUS FOR CINEMATOGRAPHIC OBSERVATION OF HUMAN CAPILLARIES

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(Received for publication February 19, 1926)

Lombard (1) in 1911 showed that by illuminating the skin the capillaries could be seen under the microscope. Since then many observations have been carried out on the changes which take place in these vessels in health and disease. The early observations which were made recorded the impressions of the author but had no quantitative basis. In recent years photographic records have been made by some investigators in order to study the changes which take place in human capillaries. This method is suitable for recording gross changes which last for a considerable length of time, such as comparing the form of the capillaries in different diseased conditions. Many changes which take place in the capillary circulation are however rapid and so fleeting that it seemed desirable, if they were to be placed on a quantitative basis, to obtain records at a more rapid rate than one could do by ordinary photography. It seemed possible that cinematographic records would enable one to follow these changes and subsequently analyse them in detail by measurement.

The possibility of studying alterations in the capillaries by means of cinematography first suggested itself to Krogh and Rehberg (2). They developed a method of taking photographs of the circulation in the capillaries in the tissues of *Rana temporaria*. It is naturally simpler to obtain records from the tissues of frogs than from human tissues since for this purpose a technique resembling that used in histological photomicrography suffices. In the human case, however, transmitted light cannot be used, so Lombard's method of illumination from above has been universally adopted. This method is difficult because by it contrast and intensity of light are much reduced com-

paired to these qualities obtained by transmitted light. The first photographs of human capillaries were taken by Weiss (3) in 1916. He devised an apparatus for accomplishing this end and published photographs obtained by his method in various diseased conditions. He used oblique¹ illumination and required an exposure of one to three quarters of a second to obtain satisfactory pictures. Siedentopf (Ziess) has devised an apparatus for the instantaneous photography of skin capillaries under normal conditions. In this apparatus also oblique illumination is used. More recently Sheard (4) has reported that he has been able to make photographs in rapid succession, but not cinematographic, of skin capillaries in the human subject. Here again oblique illumination was used but by momentarily increasing the intensity of the light at the time of the exposure, the exposure time was reduced to one tenth to one hundredth of a second. Callander (5) has also described a method which resembles those described above.

In the early stages of our work we also used oblique illumination and this was found to be satisfactory for ordinary photography but was not of sufficient intensity for cinematographic records. We therefore introduced a new feature in capillary microscopy, namely, vertical illumination. By this means a great conservation of light is obtained so that now it is possible to obtain satisfactory cinematographic pictures. These pictures are sharp and readily lend themselves to analysis by measurement. The purpose for which the apparatus was devised is in this manner fulfilled while in addition when the film is projected on the screen, the corpuscles can be seen to be in motion.

THE APPARATUS

The apparatus in its present form consists of five parts: (1) a bed and support for the arm, (2) a lighting system, (3) a microscope, (4) a stand for holding and adjusting the finger, and (5) a camera. Figure 1 shows the arrangement of the apparatus during an observation and figure 2 shows the optical system in more detail.

1 *The bed and arm-holder.* Carrier and Rehberg (6) have shown that changes take place in capillary pressure when the hand is held

¹ The terms oblique and vertical refer to the incidence of the beam of light to the plane of the finger.

at different levels below that of the heart. In order to obviate any changes due to this factor patients have been examined lying flat in bed with the arm extended horizontally at heart level. The arm is supported by an adjustable stand and fixed by sandbags. The height of the optical bench has been arranged so that the stand for holding the finger is on a level with the outstretched arm.

2 *The optical system* The source of illumination is a 15 ampere direct current arc lamp with copper coated carbons (fig 2, A). The

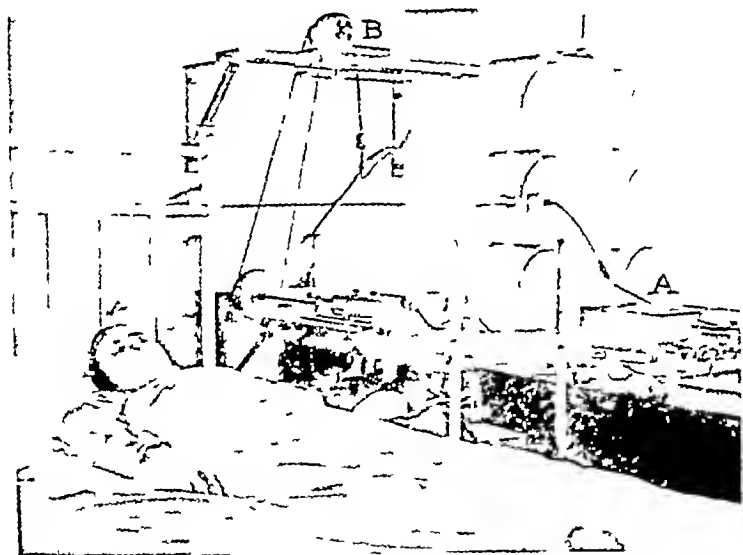


FIG 1 PHOTOGRAPH OF THE APPARATUS FOR CINEMATOGRAPHIC PHOTOMICROGRAPHY OF THE CAPILLARIES OF THE NAIL FOLD IN HUMAN SUBJECTS

intensity of the light is regulated by an adjustable resistance (fig 1, A). The light from this passes through a powerful condensing system (fig 2, BB) and also through a heat filter (fig 2, C) which is situated between the two condensers. The light is thus focussed to a small point. It enters the system for direct illumination (fig 2 D) and also passes through a polarizer. The latter has been introduced to prevent the reflection of light from the surface of the oil on the finger, which takes place with vertical illumination and prevents the capillaries

from being seen. The polarizer (fig 2, *E*) is attached to the receiving tube of the system for vertical illumination and the analyzer is situated in the tube of the microscope. With this illumination we have been able to obtain detail in our pictures to a degree which was impossible with oblique illumination. In order to learn whether the heat generated by the lighting system produced an increase in temperature at the finger we have made observations by placing a thermometer

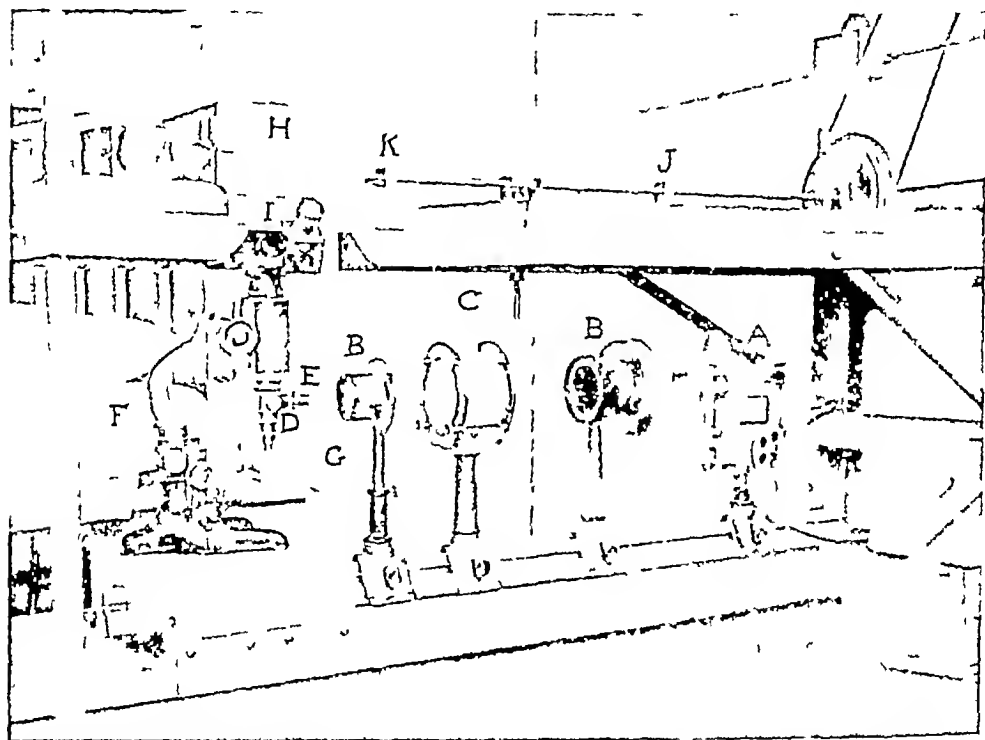


FIG 2 PHOTOGRAPH SHOWING DETAILS OF THE APPARATUS FOR CINEMATOGRAPHY OF THE CAPILLARIES

in its position. The technique was the same as that employed during the taking of pictures of the capillaries which is described below. The periods during which half strength and full strength of light were used were slightly in excess of those used in our longest observation. The temperature rose on the average 1.75°C during this period. The increase was practically constant in many observations. The greater part of the rise in temperature took place almost immediately, before the taking of the pictures was commenced. After the initial rise the

increase was very slight and gradual. Methods of eliminating even this small rise of temperature are under investigation.

3 *The microscope* In the microscopic system we use a Leitz metallurgic microscope (fig 2, *F*) with a moveable stage. The tube of the microscope is kept practically fixed and all focussing is managed by moving the stage of the microscope except the finest adjustment which is done by the micrometer screw on the microscope tube. A 16 mm apochromatic objective is used and no eye-piece. The tube of the microscope is as close as possible, without touching, to the stand which supports the camera. Through an opening in the stand on which the camera rests, there is a second short tube, the upper end of which fits the opening in the camera while the lower is telescoped into the tube of the microscope without touching it. This arrangement prevents the transmission to the microscope of any vibration caused by the mechanism of the camera. This system gives a magnification on the film of 14.4 times and has so far proved the most satisfactory arrangement.

4 *The finger holder* The stand for the finger (fig 2, *G*) has been constructed so that it is part of the stage of the microscope. The patient's finger is placed in a holder and fixed—without in any way interfering with the circulation in the finger—so as to avoid independent movement. Provision is made for adjustment in three directions by means of screws so that the finger can be placed in any desired position by the operator without voluntary movement on the part of the patient.

5 *The camera* (fig 2, *H*) was devised specially for the purpose of cinematographic microphotography. The essential difference between it and the ordinary cinematographic camera is that it works in a horizontal plane and not vertically. The camera is situated on a special stand immediately above the microscope. On its upper surface there is a viewfinder by means of which the capillaries can be seen through the film. It is thus possible to control focussing during an observation so that the sharpest possible image can be obtained. Another method of focussing which we have used is the attachment of a Zeiss observation side piece (fig 2, *I*) similar to that used in the Siedentopf apparatus, to the top of the tube of the microscope. In this arrangement it is provided that the distance from the prism

which reflects the light to the eye is the same as that from the prism to the film. This method was easier than the other but a considerable amount of light was lost so that the pictures were not quite so satisfactory. We have recently reduced the amount of light passing out at the side piece and increased that to the film thereby obtaining equally satisfactory pictures. With experience however one can see the capillaries distinctly by the former method so that it has been used in most of our observations. The film is driven by a motor (fig 1, *B*) which like the camera is on a separate stand and is operated by a foot switch. The speed of the motor is governed by a speedometer so that the number of exposures per second can be varied. The motor drives a shaft (fig 2, *I*) which can be connected or disconnected with the camera by means of a clutch (fig 2, *A*).

The stands which support the motor, the camera, and the optical

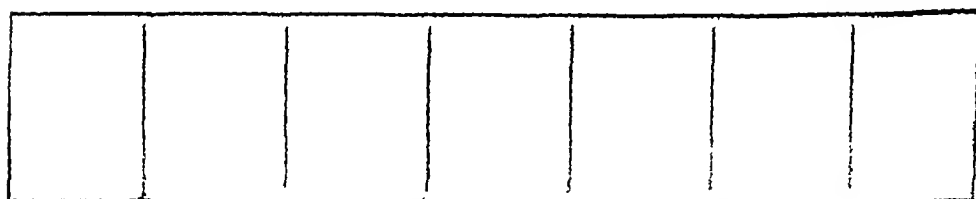


FIG 3 PRINT OF PART OF A CINEMATOGRAPHIC FILM TAKEN BY THE METHOD DESCRIBED $\times 14$

and lighting systems are in this arrangement all separate and in no way come in contact with one another. The transmission of vibrations from one to the other is consequently avoided.

TECHNIQUE DURING AN OBSERVATION

The patient lies flat in bed with the arm outstretched and the finger adjusted in the finger holder. The patient remains at rest in this position until the radial pulse has remained at a constant level for about five minutes. In cases where the pulse is irregular the observation does not begin until the variations are reduced to a minimum. Cedar wood oil is applied to the region to be examined. The light is now turned on half strength and the capillaries are brought into focus sharply on the film. The light is then turned on to the full capacity used and the motor which drives the film is started by pressing the foot

switch While the film is running the capillaries are kept under observation on the film and maintained sharply in focus by means of the fine adjustment on the microscope The radial pulse is counted throughout the entire period The temperature of the room is also noted No estimations have been made of the capillary pressure as it did not appear that these observations would be of value in the problems involved in the present series of observations

Satisfactory photographs can in this fashion be obtained (figure 3) The magnification on the film is only about 14 times but this image can be enlarged by means of a projectoscope to any desired magnification

CONCLUSION

An apparatus is described by means of which it has been possible to take cinematographic records of the capillaries at the nail fold in the human subject

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STUDIES ON HUMAN CAPILLARIES

II OBSERVATIONS ON THE CAPILLARY CIRCULATION IN NORMAL SUBJECTS

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(Received for publication February 19, 1926)

Until the later years of the nineteenth century the current view of the capillary circulation was that the capillaries were passive and that the rate of blood flow through them was determined by the state of the arteriole supplying them. Since then many researches have been carried out which show changes in the capillaries which the observers were unable to explain on the basis of arteriolar changes. The discovery by Rouget (1) of cells on the walls of the capillaries which he believed to be of a contractile nature lent strong support to the view that there is independent contractility of the capillaries. Of late years most observers agree that the capillaries both in man and in animals have the power of independent contractility. Whether they can do this normally without the application of a stimulus to them is disputed and there is also considerable doubt as to the mechanism which is involved in this action.

The literature on the subject of capillary physiology has been so admirably reviewed by Krogh (2) that only some important points in the literature of more recent date will be referred to. There seems to be no doubt that the capillaries in both man and in animals can be made to contract by the application of suitable mechanical or chemical stimuli. It has also been definitely shown in animals that electrical stimulation of nerves can cause changes in the caliber of the capillary loops independent of the changes which take place in the arterioles. The work of Krogh (2) on the muscle capillaries has demonstrated that the number of capillaries in which blood is flowing is constantly changing and that the vascular supply is adjusted to the needs of the

muscle by continuous alterations in the capillary flow Carrier (3) made observations of the skin of the forearm under the microscope and found in this situation that the capillaries were constantly changing—individual capillaries could be seen to disappear and to reappear while others appeared in new situations In their studies of the glomeruli of the frog's kidney Richards and Schmidt (4) observed that in only part of the glomeruli was blood flowing at any one time and that the capillary pathways in a single glomerulus varied No observations have been recorded in which the capillaries at the nail fold in man have been seen to appear and disappear as described by Carrier in the corresponding vessels of the forearm but changes in the flow in these vessels have been frequently described

A question which has interested many observers is whether under normal conditions independent contractility is displayed by the capillaries and whether the nature of this contraction is a peristaltic wave which would tend to propel the blood forward Variations in the flow undoubtedly take place but whether these are due to contraction in the capillaries themselves is uncertain Krogh (2) brings forward evidence in favor of the view that the changes which take place in muscle are of this nature But Richards (4) in his work on the frog's kidney has been unable to find any evidence in support of the idea that independent contractility takes place under normal conditions Many observers have stated that they have seen changes in the capillaries of the nail fold in man under the microscope which they interpret as showing evidence of capillary contractility Thaller and Draga (5) described alterations which they think to be of the nature of peristaltic contractions of the capillaries This view has been strongly upheld by Kohn (6) Krogh (2) on the other hand has seen no evidence in favor of the occurrence of this mechanism in his observations in human subjects and thinks the conception is probably incorrect Muller (7) as the result of extensive studies in the Tubingen school agrees with Krogh and does not think that the changes can be due to peristalsis Many observers have brought forward evidence both for and against this view

Although the conclusion that capillaries can contract independently has been generally accepted, two views are held as to the cells which are responsible for this behavior Some believe that it is caused by

changes in the endothelial cells as first suggested by Stricker (8) and others that it is due to cells outside the endothelium of the capillary wall. In 1873 Rouget (1) described cells in this position which he thought to be of the nature of smooth muscle and to possess a contractile function. The existence of these cells has been amply confirmed. Under Krogh's direction Vimtrup (9) has carried out an extensive study of their nature chiefly by histological means. In his report of these studies Vimtrup gives a full review of the literature of this phase of the subject. He confirms Rouget's results and says that he has been able to trace the transition from the smooth muscle of the smallest arteries to the Rouget cells. He also studied the changes which take place in the capillary wall in the living animal and states that the contraction which follows nerve stimulation always starts at one of these cells. Clark and Clark (10) have studied the problem from a different angle. They found it possible to observe the development of the capillaries in the tail of living amphibian larvae and to make camera lucida drawings of the selected area under observation at frequent intervals for eight days. They found that the Rouget cells were not contractile elements but developed from stellate connective tissue cells. In their opinion two types of contraction occur in the capillaries, an active one which may occur spontaneously, and a passive one which is caused by temporary disturbance of the circulation. Both types of contraction were independent of the presence of adventitial cells. Contraction was indeed often to be seen before the development of these cells. After they were developed contraction began frequently at places at a distance from them. During active contraction or passive narrowing of the vessel, the capillary might be seen to draw away from the Rouget cells and to leave a space between cell and wall when viewed from a suitable angle. They came to the conclusion that both the power of contractility and tonicity are independent of any form of adventitial cell.

The present observations were undertaken in order to study the nature of the changes which take place in the capillaries by means of cinematographic records. Exposures were taken at intervals of $\frac{1}{16}$ second. It was possible therefore to measure the diameter of the capillary walls throughout a long series of consecutive photographs with the hope of throwing light on certain of the problems which have

been discussed above. The technique of making the photographs has been described in another paper (11). The results reported are based on a study of the capillaries at the nail fold in 8 normal individuals (4 men and 4 women) between the ages of 25 and 35. Observations on all cases were made on 4 consecutive days at the same hour. In every instance the temperature of the room showed practically no change from day to day.

METHOD OF STUDYING THE FILMS

Projection In the film the individual capillaries were magnified 14.4 times. With this magnification it would be impossible to estimate any but gross changes with any degree of certainty as the error in measurement would be so great. The greater the magnification at which the contour of the capillary was sharp the less the error in measurement. A special projectoscope which enlarged the capillary 350 times was constructed thus considerably reducing the possibility of error. At this magnification a sharp picture of the capillary was obtained. The contour of the individual capillaries in a large series of consecutive pictures was traced on paper and these tracings were used for measurement.

Measurement The point at which measurement is made is of paramount importance. The most satisfactory arrangement would be to provide a base line in the tissues and to measure the diameters of the arterial and venous limbs at a constant distance from this line. It was not however practicable to do this. A marker placed on the finger might also have been tried but its position was liable to be fallacious as it might not follow accurately any movement of the finger itself which took place. An indicator close to the film might lead to inferences even more fallacious. In place of methods of this sort we have selected a point on the magnified image 2 cm. from the tip of the capillary loop (0.06 mm. from the tip of the actual capillary) as the point at which to measure the diameters of the arterial and venous limbs. The tip of the loop is taken as the point of reference, for, barring one reservation, movement here is accompanied by movement of other points in the loop. Between the tip and any other nearby point on the loop, the distance may be regarded as remaining constant. The reservation which was mentioned concerns the possibility of

lengthening of the loop for any reason, notably that which may occur at the time of cardiac systole. The method of measuring was as follows. A line was drawn through the center of the long axis of the capillary loop roughly parallel with the two limbs and at right angles to this a second line which touched the tip of the loop. A third line was drawn parallel to the second at a distance of 2 cm. which cut the arterial and venous limbs (fig 1). Changes in the diameters of these limbs at this level have been taken to indicate changes in behavior of the capillaries. Curves were prepared in which the values so obtained were charted, the abscissa representing the time and the ordinate the diameter of the limb. These have been used for comparison of the changes in the various observations. In other observations of the same subject other methods of measurement were used and the several curves compared to those obtained by the method just described. The opportunity for comparison occurred in certain capillaries in which owing to their peculiar shape two or more characteristic points could be selected as reference points (fig 1). In such cases parallel lines were drawn through the points selected and at right angles to the long axis of the loop. The distance between these was measured.

Obviously several interpretations may be drawn of the behavior of the capillaries, depending on changes which are disclosed when the measurements are taken. 1. If the distance ($a-b$) remains uniform and the measurements of the diameter (c) at a point 2.0 cm. from the tip of the loop show no significant variations, it is unlikely that an important change in the state of the capillary has taken place. 2. If the distance ($a-b$) increases, and the measurements of the diameter (c) decrease, the inference may be drawn that stretching of the capillaries has taken place. If on the other hand the measurements of the diameter (c) increase or remain unchanged, an effect on the vessels by stretching may be excluded. The change in measurement is then the result of another function. 3. If the distance ($a-b$) decreases, and the measurements of the diameter (c) increase, the inference may be drawn that this result depends on the decreased length of the tube. But if the diameters (c) decrease or remain unchanged an effect on the vessels as a result of this action may be excluded and must depend then on another function. In point of fact, behavior of all these types was observed. It was impossible to infer that uniform changes

in the diameters took place. From these observations one cannot draw a conclusion on the events which take place in the capillaries during the heart beat. They permit the statement to be made how-

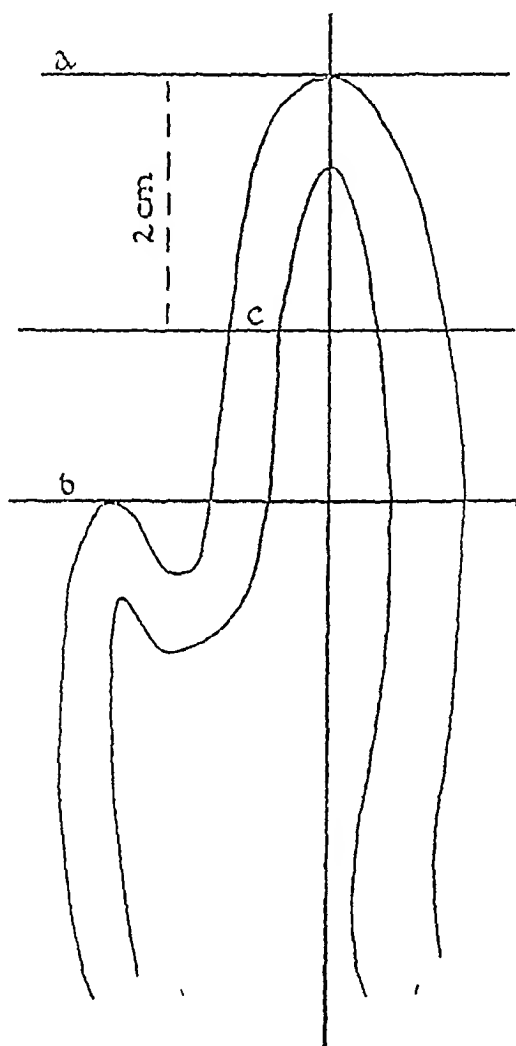


FIG. 1 SCHEMATIC DRAWING OF A CAPILLARY TO ILLUSTRATE THE METHODS WHICH WERE EMPLOYED IN THE MEASUREMENT OF THE DIAMETERS OF THE LIMBS OF THE CAPILLARY LOOPS

(For description, see text)

ever that changes in the diameter, should they be found to occur, do not depend on this factor. They do moreover show that to measure the diameters 2.0 cm from the tip exposes the measurements to no

systematic distortion To obtain this measurement was our purpose Curves drawn from observations of capillaries which afforded this opportunity did not differ in form from those obtained when it was impossible to utilize this check

In many of the films photographs have been obtained of 4 or more capillaries all of which were in sufficiently sharp focus to allow accurate measurements The positions of the tips of all these loops have been compared with respect to one another in a large series of exposures over a long period of time It was found that these remained constant But the objection may be made that the tips of all these changed at an equal rate, and that therefore no relative changes could be observed When adjacent loops were compared by measurement of the diameter it was found that the various loops showed changes which were quite dissimilar In other words, no relative movement of the tips takes place or should there be movement, this is congruous whereas the measurements of the diameters are incongruous The total area of the loop distal to the line drawn 2 cm from the tip through the arterial and venous limbs has also been measured by means of the planimeter and it was found that this area remained practically constant As a result of a comparison of these measurements the method first described appears to be subject to slight error only and provides a useful way of estimating the changes which take place in the loops

OBSERVATIONS

The size of the loops As is well known there is great variation in the size of the capillary loops at the nail fold both in the same subject and in different subjects It seems almost useless to state the size of the normal capillary as the variations between capillaries in the same subject and in different subjects is so great A review of the literature shows that it is not possible to compare the values given by different authors unless these are measured at the same point on the loop The measurement of the arterial limb in most of the present observations at the selected point has varied from 0.014 to 0.016 mm while that of the venous has usually been slightly larger, 0.015 to 0.017 mm In many instances, however, smaller and larger capillaries have been measured but similar variations have taken place in all

Variations in diameter Both the arterial and venous limbs of the same capillary varied in diameter from moment to moment and from day to day in the same subject (fig 2) Although these changes take place they are relatively small compared to the breadth of the limbs

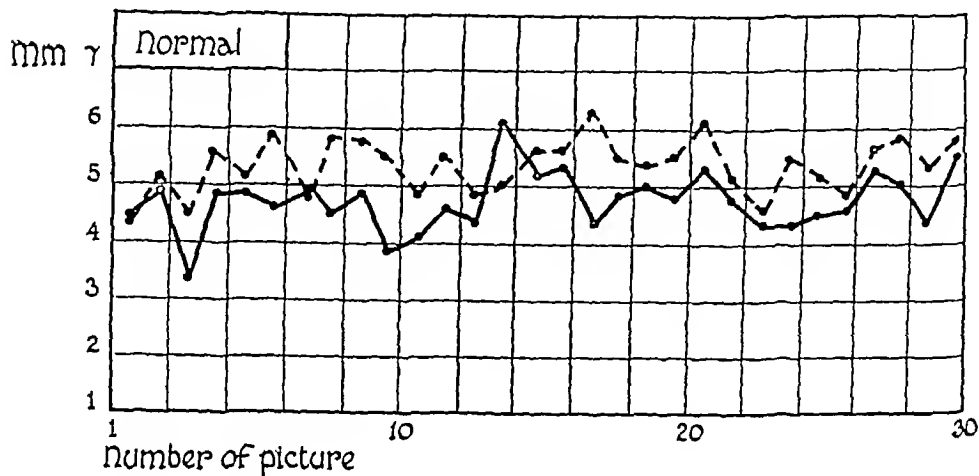


FIG 2 CHANGES IN CONSECUTIVE PICTURES IN THE DIAMETERS OF THE ARTERIAL AND VENOUS LIMBS OF A CAPILLARY IN A NORMAL SUBJECT

Abcissa = number of the picture There were 10 exposures per second
 Ordinate = diameter in millimeters at a point 2 cm from the tip after magnification of the capillary 350 times • — — • arterial • — — • venous

Normal

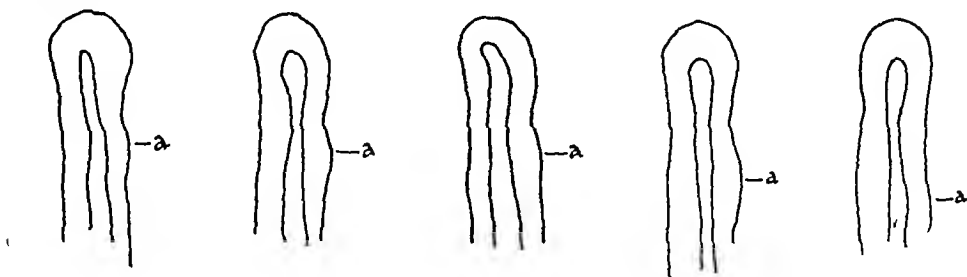


FIG 3 TRACINGS OF FIVE CONSECUTIVE PICTURES OF A CAPILLARY LOOP IN A NORMAL SUBJECT $\times 350$

which remains approximately the same from day to day These changes are very noticeable on observation of successive magnified pictures (fig 3) The daily changes were of a similar nature to those taking place from moment to moment while the extent of the varia-

tions was usually about the same in the two limbs. The magnitude of the changes also varied from capillary to capillary and from subject to subject. In some it was small while in others it was much more marked.

Evidence of a peristaltic wave of contraction If the mechanism which is responsible for the changes which take place were peristaltic in nature recurrent alterations in form ought to occur with approximate regularity as regards both time and magnitude. We have studied all our curves from this point of view but have been unable to find any evidence for the presence of this mechanism (fig 4). In a few in-

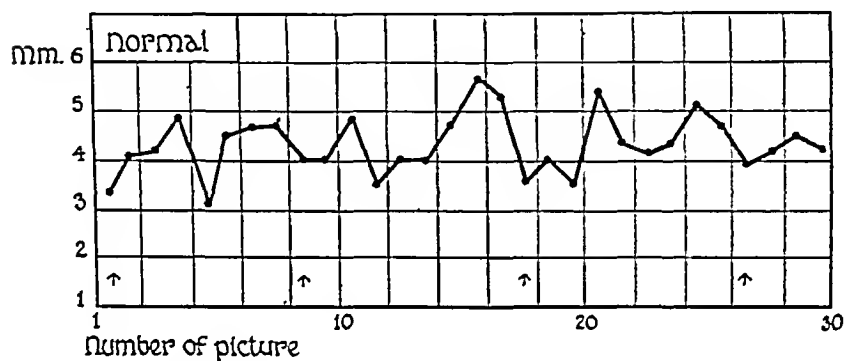


FIG 4 CHANGES IN THE DIAMETER OF THE ARTERIAL LIMB OF A CAPILLARY IN A NORMAL SUBJECT $\times 350$

↑ ——— ↑ = one complete cardiac cycle

stances a small part of the tracing suggested this possibility but the subsequent changes through a long period of observation show that this occurrence was merely accidental. One of these rare instances is illustrated (fig 3). A hump (*a*) can be seen on the loop and this can be followed throughout the series of pictures. If this hump were due to a peristaltic wave it might be expected to proceed in the same direction at a constant speed. The most prominent point on the hump has been measured with reference to a line drawn through the tip of the capillary and it was found that both the direction and speed varied. In the second picture it was slightly lower, in the third it had returned to practically its original level while in the fourth and fifth it became

progressively lower. A large number of consecutive pictures studied later in the series showed no evidence of a hump in this situation.

Evidence of rhythmical contraction of the capillaries A second type of motion of the capillaries is possible, namely, rhythmical contraction. Observations on this question have been made by extended inspection of the capillaries themselves and the curves of the measurements have also been studied. In neither method has any evidence in favor of this mechanism been seen.

Evidence of changes of a pulsatory nature In the examination which was next undertaken it was the object to discover whether the changes that have been described depend on the cardiac pulse. In this case again, the phenomenon should be recurrent. Knowing the heart rate and the number of exposures per second, photographs corresponding to the same phase of the cardiac cycle can be measured and compared. If the behavior of the capillary were congruent with the pulse, the measurements of corresponding photographs should be similar and should form a characteristic curve. It has been impossible in any of our tracings to relate the changes to a mechanism of this nature (fig 4). In some cases the diameter of the capillary remained approximately the same for the duration of a heart beat while in the following heart beat for the same period it was either considerably larger or smaller. In most cases however the changes were totally irregular.

Blood flow The blood flow was observed by simple inspection of the capillaries with the microscope but no measurements of this phenomenon were made. The blood flow varied in the different capillaries in the same subject and also in the same capillary from moment to moment. At one time the flow might be very rapid while a little later the stream might become very sluggish and indeed might stop momentarily. All gradations between these two extremes were to be observed but in this situation capillaries have not been seen to disappear and others appear, as has been described in other situations. A considerable difference could also be observed in the capillary circulation of the different subjects studied.

DISCUSSION

The observations which have been made show that the diameter of the capillaries at the nail fold in man is constantly changing. Al-

though individual capillaries do not appear and disappear as they do in certain other tissues nevertheless definite variations occur in this situation. They also show that the reactions which take place are of a similar nature over a period of several days and that different subjects exhibit individual peculiarities in the changes seen. These have been described by other observers but we have now been able to measure them and to record the rapidity with which they can take place. These are discussed later in detail.

In these observations as in all similar studies in man the diameter of a capillary limb has been inferred from the breadth of the stream of blood flowing in it as the wall of the vessel cannot be identified. In order to make accurate statements one ought to be able to photograph the actual wall of the capillary which is impossible. The width of the blood stream has been used as a criterion because the capillaries remain continuously full of blood except for the occurrence occasionally of small gaps in the stream whose significance is discussed below. A diligent search has been made by inspection to discover whether there is evidence of an axial stream of corpuscles and a peripheral zone of pure plasma in the capillaries. If this arrangement were found the corpuscular stream would not be an accurate representation of the changes in the capillary wall. Observations made on this point have confirmed those of Krogh (2) that this zone of pure plasma is practically non-existent. We have concluded therefore that the breadth of the corpuscular stream offers a basis for measuring the diameter of the capillary loop in this situation and can be used as such.

It is particularly important in a study of the circulation to know whether under normal circumstances the capillaries play an active part in the forward propulsion of the blood. Changes have been seen which have been taken to be movements in the walls of the capillaries and an endeavor has been made to relate this to the occurrence of certain possible rhythmical mechanisms. There are two types of motion of which the capillaries might, as has been said, be capable. The first is peristaltic contraction and the second local rhythmical contraction. It has been impossible to correlate either of these mechanisms with the changes observed. The former has received considerable support based principally on the fact that gaps in the stream

of corpuscles have been observed. These gaps are not often seen in normal subjects and usually take place when the stream is slow. Krogh (2) discussed the significance of these gaps and did not consider that they were due to peristaltic contraction of the capillary wall. One can watch many capillaries for a long time without these gaps appearing and in those in which they are seen they appear so irregularly and unless the stream is very slow so infrequently that they do not appear to be due to a peristaltic wave. The rate of progress of the gaps is very variable and often very rapid and the whole appearance suggests a gap in the corpuscular stream rather than a contraction of the wall. The mechanism involved is probably an agglutination of corpuscles which causes a momentary stoppage of blood flow and thus allows a gap to be formed.

The cause of the variations which were found is in doubt. They may be due to local contraction of the capillary wall occurring in an irregular manner. Or they may be due to alterations in the tonicity of the arterioles. It has been definitely established that the latter possess tone (12) and frequent spontaneous changes in their tonicity, controlled in great part by the central nervous system, have been shown to take place by Langley (13). There is however an objection in relating what has been seen in the capillaries to the contraction of the arteriole as a whole, for if this were the responsible agent all the capillaries supplied by an arteriole should be similarly affected. But as has been shown, similarity of action on the part of the capillaries in the same field is not the case. Tarchanoff (14) has found however that on applying an electrical stimulus to the capillaries of a frog's nictating membrane contraction appeared much sooner at the junction of the arteriole and capillary than in the rest of the capillary. Richards (4) states that experiments in his laboratory on arterioles and capillaries in the muscle of the frog showed that the action of adrenalin is more marked at the branching of the vessels than along their course. He thinks that this may be the explanation of the variations in flow which are observed in the frog's kidney. The changes seen in the present studies might thus be related to variations in the tonicity of the arteriole at its junction with the capillary. Which of the possible explanations, capillary contraction or changes in arteriolar tone, is correct, remains unsolved by these investigations.

It appears then that the capillaries in the nail fold behave differently from those located in other tissues of the body, at least in other species than man. For in man the capillaries of the skin and visible mucous membranes only seem to have been examined, in animals there is, of course, information of their motions also in certain viscera. It is of some importance to keep in mind this phase of the state of knowledge of this subject. In this study there has been added a precise description of what was observed in a single portion of the skin. To remember this has its importance in view of the difference in action which Carner has seen in the capillaries of the skin of the forearm. It may be said in a general way then that a single capillary area does not supply the criteria which may serve as a basis for making statements of the mechanism of the capillaries in the whole body. It is necessary apparently in making calculations of the circulation and in dealing with other physiological processes such as respiration and edema to allow for this matter of diversity of function.

CONCLUSIONS

1 The caliber of the arterial and venous limbs of the capillaries at the nail fold has been studied by means of cinematography over a period of days in 8 normal individuals.

2 Changes took place from moment to moment and from day to day. These changes were relatively small compared to the total breadth of the limbs which remained approximately the same from day to day.

3 The extent of the changes in the arterial and venous limbs of the same capillary was about the same, but the various capillaries in the same subject different from one another. Marked variations were also seen between the different subjects. There was no evidence that these changes were due to a peristaltic wave of contraction, a contractile motion of the capillary like that of the heart, or a pulsatile motion conveyed to the blood stream by the heart beat. The mechanism of their production is uncertain.

4 The blood flow in these capillaries has been studied by inspection and was seen to vary continuously.

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STUDIES ON HUMAN CAPILLARIES

III OBSERVATIONS IN CASES OF AURICULAR FIBRILLATION

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(Received for publication February 19, 1926)

In a study of the adjustment of the circulatory mechanism to different abnormal conditions in the heart the changes induced by circulatory failure are of special interest. As auricular fibrillation is an important cause of heart failure the study of cases of this nature is particularly important.

The first microscopic studies of the capillaries in living subjects in heart disease were made by Weiss (1) who found that in cases with good compensation the capillaries resembled those of normal individuals. In the early stages of mitral stenosis however a distinct slowing of the stream was often observed while in some cases of pulmonary stenosis there was very marked stasis. Advanced cases of cardiac decompensation showed marked dilatation of the venous limb, a tendency to tortuosity of the capillaries and a dark red color of the tissues in which the capillary loops were situated. The color of the latter varies with the intensity of color of the blood flowing in the deeper lying vessels. The blood stream was slow and discontinuous and the flow had a granular appearance. The subpapillary plexus was more distinct than in normal subjects. These findings were confirmed by Schur (2), Jurgensen (3), Neumann (4), Secher (5), Friedlander and Lenhart (6). Secher (5) stated that early cases of mitral stenosis showed a normal picture. Marked stasis was described by Rominger (7) in a case of pulmonary stenosis. Hisinger-Jagerskiöld (8) carried out an extensive study in cases of heart disease of valvular and myocardial origin in some of which the pulse was regular and in others irregular. He stated

that cases with full compensation or with only slight decompensation presented a normal picture whereas in decompensated cases changes took place. Those cases which exhibited edema or cyanosis of the periphery showed changes of the sort associated by Weiss with marked circulatory failure. He differentiated a group of cases in which these phenomena were absent although there was marked congestion of the internal organs. In such cases the color of the loops was normal and the background light red. Some of the loops were smaller than normal and all the loops were well filled. The stream was sometimes rapid and sometimes slow. This picture resembled that which the author had observed in cases of anemia. The various changes seen were independent of the character of the pulse and altered with the state of compensation. In his opinion the different pictures were dependent on changes of blood volume.

The results to be reported are based on the study of seven cases of auricular fibrillation of rheumatic origin except one which was associated with hyperthyroidism. Cinematographic observations of the capillaries at the nail fold were made in these cases at frequent intervals over a prolonged period. Cardiac decompensation was well marked in all but its severity varied. Observations were made during the period when patients were not under the influence of digitalis and also during the course of digitalis administration until full digitalization was established. Details of the technique of taking the photographs and the methods employed in their study have been reported in previous papers of this series (9, 10). As the rate of the pulse in auricular fibrillation is very unstable patients remained at rest in quiet surroundings until variations in the pulse rate were reduced to a minimum and remained practically constant for a considerable period.

The main interests in the present investigation were to study whether the size of the loops and the variations in their caliber varied from those which had been observed in the normal individuals which had been investigated, whether there was any evidence of contractility of the capillaries themselves or whether changes of a pulsatory nature due to the heart beat were present, whether the blood flow varied from that seen in normal individuals, and finally whether any changes observed were altered by digitalization.

OBSERVATIONS

The size of the loops A marked variation in the size of the loops was present in all cases. It differed in the different subjects and was also markedly altered by the state of decompensation. The milder degrees of decompensation showed a picture closely resembling normal but the loops were usually definitely enlarged in those cases which showed severe circulatory failure. The venous limb of the loop was more affected than the arterial although in the more severe grades this was also involved. The size of the loops was so variable that one is unable to give figures of any value of their size. The caliber of the arterial limb at the point selected for measurement averaged 0.016 mm in most of the loops measured while that of the venous was 0.018 mm. In many instances larger and smaller capillaries were measured but the variations which are described below were of the same nature in all.

Variations in caliber Changes in caliber took place in the same subject from moment to moment in both the arterial and venous limbs to about an equal degree (figs 1 and 2). No two capillaries were alike in the time and extent of these changes and individual differences were evident between the various subjects studied. Although these changes were present the total breadth of the limbs remained practically the same from day to day. Similar changes of caliber had been observed in the capillaries of normal subjects but those seen in auricular fibrillation were more extensive. The degree of decompensation also had a pronounced influence, the more marked the decompensation the more marked the variations in caliber.

Evidence of independent contractility of the capillaries The curves which were prepared in the manner previously described have been carefully studied to see whether (1) a peristaltic wave of the capillaries or (2) local rhythmical contractions similar to those of the heart itself were present. It seemed that if the "peripheral heart" really existed its action might play an important part in the circulatory mechanism in heart failure. No evidence has been found in favor of these hypotheses as the changes took place irregularly and showed no tendency to rhythmicity such as ought to be present if these factors were operative (fig 3).

Evidences of pulsation due to the heart beat In auricular fibrillation

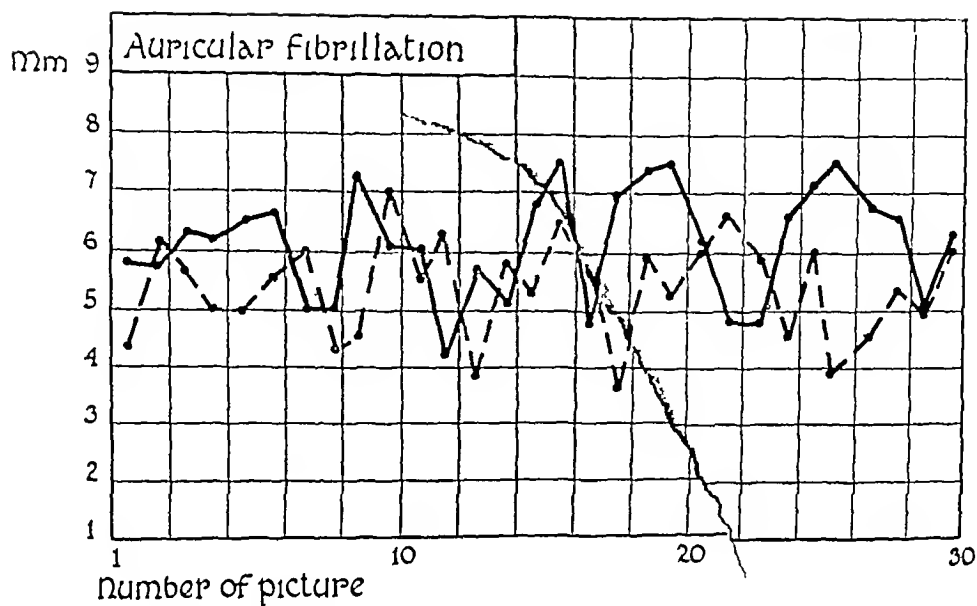


FIG 1 CHANGES IN CONSECUTIVE PICTURES IN THE DIAMETER OF THE ARTERIAL AND VENOUS LIMBS OF A CAPILLARY IN A CASE OF AURICULAR FIBRILLATION $\times 350$

• — • arterial • - - • venous

Auricular fibrillation

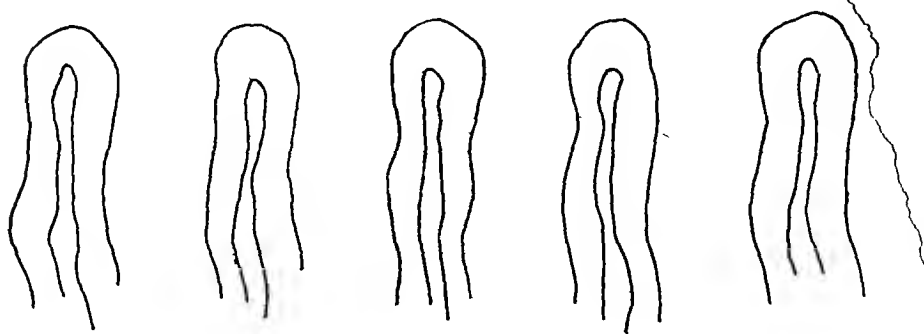


FIG 2 TRACINGS OF FIVE CONSECUTIVE PICTURES OF A CAPILLARY LOOP IN A CASE OF AURICULAR FIBRILLATION $\times 350$

it is much more difficult than in cases with normal rhythm to be certain that changes which take place irregularly are not induced by cardiac contraction which forces blood into the small vessels as in the former condition the cardiac contractions themselves vary in time and force. This would apply particularly in those cases in which the heart was very irregular both in time and force. In a few cases marked decompensation was present but the ventricular rate was slow and the irregularity in the time and force of the heart beats was slight. One would expect that if pulsation in the capillaries due to the irregular

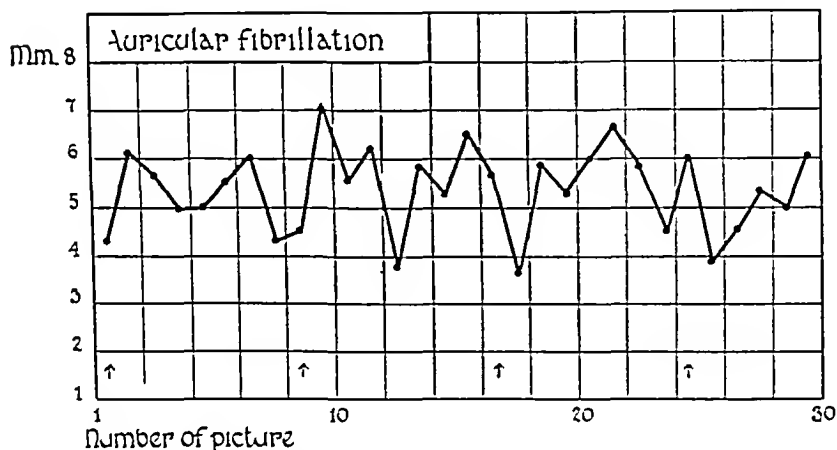


FIG 3 CHANGES IN THE DIAMETER OF THE ARTERIAL LIMB OF A CAPILLARY IN A CASE OF AURICULAR FIBRILLATION WITH A SLOW VENTRICULAR RATE $\times 350$

↑ ——— ↑ the approximate duration of each cardiac cycle

heart beat observed in auricular fibrillation were the mechanism involved the cases which showed the least irregularity of the cardiac contraction would show less violent fluctuation in the capillary changes. No difference could be detected as whatever changes took place were indeed equally irregular in both. As further evidence against this being the mechanism present two cases of marked cardiac decompensation with regular, rather than irregular, rhythm might be cited in which like changes were seen to those in cases of auricular fibrillation with a comparable degree of decompensation (fig 4)

Blood flow The blood flow was studied by inspection over prolonged periods. The rate of flow was continuously changing in the same capillary from moment to moment. All the capillaries in the

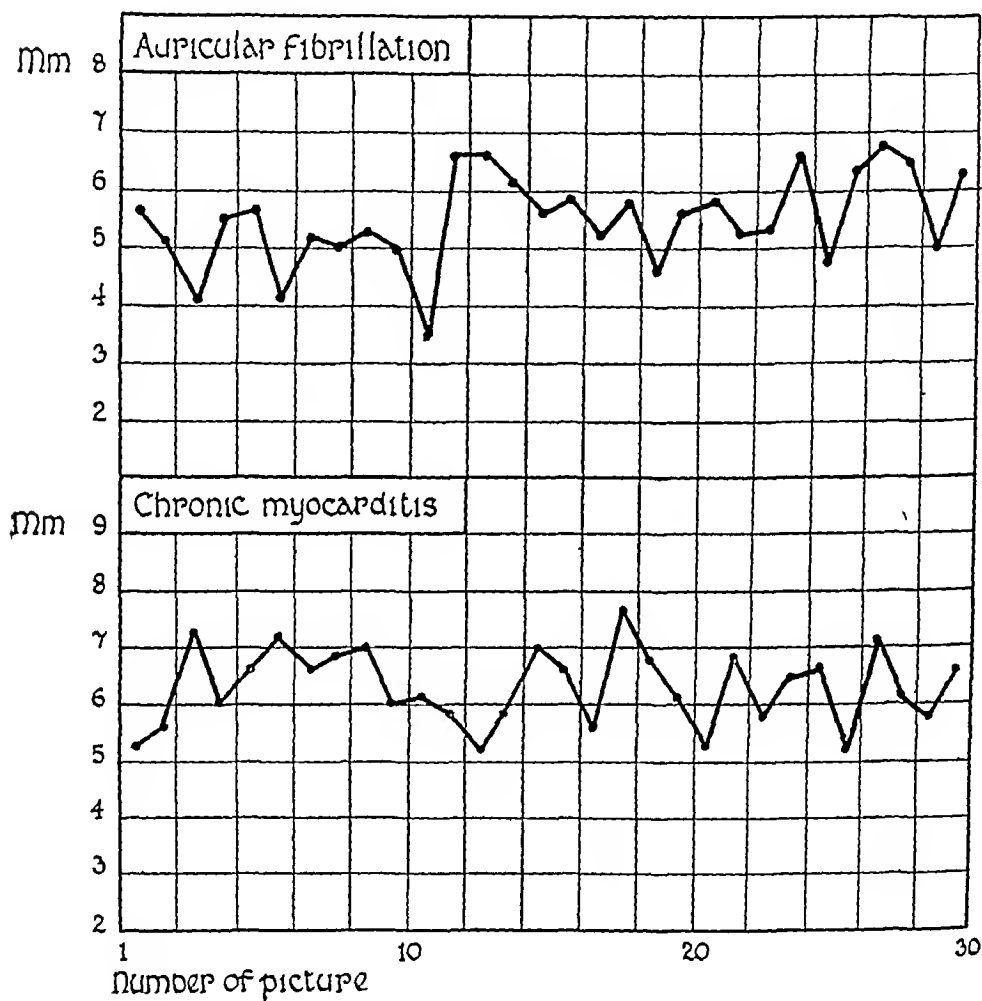


FIG 4 CHANGES IN THE DIAMETER OF THE ARTERIAL LIMB OF A CAPILLARY IN A CASE OF AURICULAR FIBRILLATION AND SIMILAR CHANGES IN A CASE OF CHRONIC MYOCARDITIS WITH REGULAR RHYTHM WHICH SHOWED A COMPARABLE DEGREE OF CARDIAC DECOMPENSATION $\times 350$

same field differed from one another. In most capillaries the stream was much slower than had been observed in normal individuals. The slowing of the stream varied with the degree of decompensation. In the most marked cases the stream in almost all the capillaries was very

slow and in many stasis was present which often lasted for a considerable time. The flow was very irregular, a capillary might show at one moment stasis and the next moment the stream in it might be very rapid. Gaps in the corpuscular stream were often seen giving the granular appearance of the stream which has been described by pre-

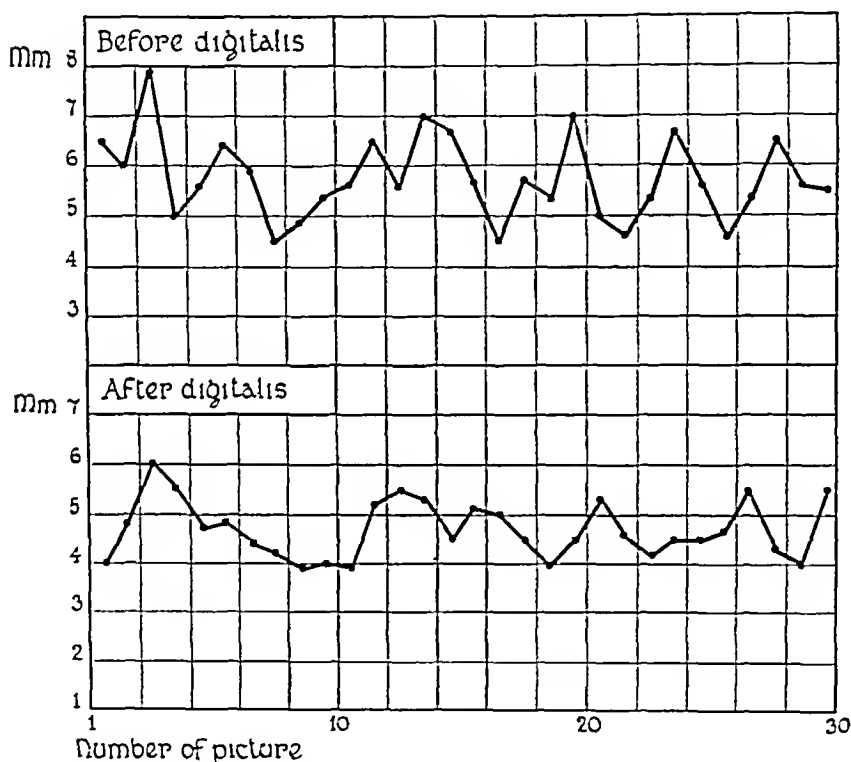


FIG 5 CHANGES IN THE DIAMETER OF THE ARTERIAL LIMB OF A CAPILLARY IN A CASE OF AURICULAR FIBRILLATION BEFORE AND AFTER DIGITALIZATION $\times 350$

vious authors. In some cases a backward flow from the venous to the arterial limb was momentarily present.

Digitalis All cases have been studied daily during the period at which they were coming under the influence of digitalis. Most of the cases as the result of digitalization showed definite changes in the

capillary circulation which varied with the extent to which the efficiency of the circulation was improved. Very little difference was seen in the average breadth of the arterial and venous limbs. When the venous limb was very large, however, before digitalization it was sometimes reduced in size to a slight extent. The extent of the variations in caliber which have been described was reduced in all those cases which were improved by digitalis (fig 5). In a subject in whom full compensation was established the changes approximated closely to those observed in normal individuals. The blood flow improved in a corresponding manner. The rate of flow in the individual capillaries became more nearly similar, the frequency and duration of stasis were reduced and a granular appearance of the stream was less frequently present.

DISCUSSION

The variations which took place in the caliber of the capillaries in auricular fibrillation were of the same nature as those which took place in normal subjects except that the magnitude of these changes was increased. The degree to which this was so did not depend on the heart rate or on the irregularity of the pulse. It appeared to be dependent solely on the extent to which the circulation was inefficient as judged by other, more general, criteria. This was shown clearly by the correspondence between the extent of the changes and the clinical condition of the patient in cases in which the rate and rhythm of the pulse remained practically unaltered. When the state of decompensation was relieved by digitalis a lessening of the variations in caliber took place. The type of change in the subjects in which compensation was fully established approximated closely to normal.

The blood flow in the capillaries in cases of cardiac decompensation presents a striking picture. The circulation is known to be inefficient but a beautiful demonstration of the extent to which this is so is given by the observation of the flow in these small vessels. In a marked case stasis may be present in a majority of the capillaries all the time although not permanently in the same capillary. The correspondence between the improvement of the blood flow and the subjective symptoms of the patient is striking.

As is well known capillaries can contract under suitable stimuli

It was thought possible that the existence of the state of heart failure might provide such a stimulus. Previous observations in the present series had given no evidence of independent contractility of the capillaries in normal subjects (10). Nor has any evidence been obtained of such a mechanism in cases of heart failure.

The explanation of the changes is in doubt. If they are due to active contraction of the capillaries which seems to us most improbable, these contractions must take place in a very irregular manner. The probability is that the mechanism involved in their production is the same in both normal individuals and in patients with cardiac decompensation. The possible factors involved in normal subjects has previously been discussed (10). Whatever difference exists between the behavior of the capillaries in normal individuals and in cases of auricular fibrillation is probably to be accounted for by the inefficiency of the heart itself.

CONCLUSIONS

1 The caliber of the arterial and venous limbs of the capillaries at the nail fold has been studied by means of cinematography in seven cases of auricular fibrillation both before and after digitalization.

2 Changes of caliber of about equal magnitude took place in the arterial and venous limbs from moment to moment. The behavior of the various capillaries in the same subject differed from one another. The extent of the changes during the stage of decompensation was much greater than had been observed in normal subjects but as compensation became established under digitalis the variations became less marked.

3 The magnitude of these changes was dependent on the state of cardiac decompensation and had no relation to the rate or amount of irregularity of the pulse.

4 There was no evidence that these changes were due to a peristaltic wave of contraction, a local rhythmic contractile action of the capillary or a pulsatile motion conveyed to the blood stream and so to the capillary wall by the heart beat. The mechanism of their production is uncertain.

5 The blood flow in the capillaries has been studied by inspection. The rate apparently varied from subject to subject and from capillary

to capillary in the same subject In cases of marked decompensation the stream was slow in most of the capillaries and had a granular appearance while in many, stasis was present As compensation became established there was a corresponding improvement in the blood flow

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JAUNDICE FOLLOWING PULMONARY INFARCTION IN PATIENTS WITH MYOCARDIAL INSUFFICIENCY

I A CLINICAL STUDY

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(Received for publication January 30, 1926)

INTRODUCTION

Jaundice occurring in myocardial insufficiency is a well recognized condition. It is seen most frequently in patients with long standing heart failure, particularly in patients with mitral or tricuspid disease and auricular fibrillation. The studies of van den Bergh and Snapper (1913), Lepehne (1921), Eppinger (1923), Feigl and Querner (1919), McNee (1922-23), Fishberg (1923), Andrews (1924), and others, have shown that jaundice in myocardial insufficiency is due to an increase of the bilirubin content of the blood serum. The subject we wish to discuss is the sudden appearance of jaundice or the marked increase in the jaundice, which occurs after pulmonary infarction in patients suffering from myocardial insufficiency.

The only references in the literature that we have been able to find in which a specific relationship between jaundice and pulmonary infarction has been suggested have been those of Schottmuller (1914), Eppinger (1923), and Libman (1923). There are many other instances in which pronounced jaundice has been associated with pulmonary infarction, but the significance of the association has evidently not been appreciated, at least, it was not mentioned or discussed. Among those are reports by Feigl and Querner (1919), Oertel (1910), and Mann (1907). Of especial interest in this regard is the statement of Matthes (1925), that one of the characteristic features of jaundice in patients with myocardial failure, is its sudden appearance, although no reference is made to the possible dependence of the jaundice on pulmonary infarction.

to capillary in the same subject In cases of marked decompensation the stream was slow in most of the capillaries and had a granular appearance while in many, stasis was present As compensation became established there was a corresponding improvement in the blood flow

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JAUNDICE FOLLOWING PULMONARY INFARCTION IN PATIENTS WITH MYOCARDIAL INSUFFICIENCY

I A CLINICAL STUDY

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(Received for publication January 30, 1926)

INTRODUCTION

Jaundice occurring in myocardial insufficiency is a well recognized condition. It is seen most frequently in patients with long standing heart failure, particularly in patients with mitral or tricuspid disease and auricular fibrillation. The studies of van den Bergh and Snapper (1913), Lepehne (1921), Eppinger (1923), Feigl and Querner (1919), McNee (1922-23), Fishberg (1923), Andrews (1924), and others, have shown that jaundice in myocardial insufficiency is due to an increase of the bilirubin content of the blood serum. The subject we wish to discuss is the sudden appearance of jaundice or the marked increase in the jaundice, which occurs after pulmonary infarction in patients suffering from myocardial insufficiency.

The only references in the literature that we have been able to find in which a specific relationship between jaundice and pulmonary infarction has been suggested have been those of Schottmuller (1914), Eppinger (1923), and Libman (1923). There are many other instances in which pronounced jaundice has been associated with pulmonary infarction, but the significance of the association has evidently not been appreciated, at least, it was not mentioned or discussed. Among those are reports by Feigl and Querner (1919), Oertel (1910), and Mann (1907). Of especial interest in this regard is the statement of Matthes (1925), that one of the characteristic features of jaundice in patients with myocardial failure, is its sudden appearance, although no reference is made to the possible dependence of the jaundice on pulmonary infarction.

Schottmuller (1914) recorded a case in which jaundice occurred after a pulmonary infarction and he felt that the jaundice was due to the presence of hematin in the serum. However, he did not state whether or not the patient had myocardial insufficiency or whether he had an increased bilirubinemia as well.

There are other instances in patients with myocardial insufficiency in which jaundice may *seem* to appear suddenly. These instances occur most frequently during convalescence or during a period of marked improvement. This condition is not to be confused with the sudden appearance of jaundice that occurs after pulmonary infarction, because it is not at all unusual for the jaundice in cardiac patients to become more distinct when the cyanosis disappears, since the cyanosis tends to mask the icterus. Another explanation for this phenomenon may be the wide fluctuation of the bilirubin content of the blood plasma which may occur from day to day even while the patient is improving. This type of jaundice may be easily differentiated from the type that appears following pulmonary infarction since, in the one case the patient continues to improve, while in the other he usually becomes progressively worse.

We wish to report in this first communication, ten cases in which jaundice appeared for the first time, or became more intense, after the occurrence of pulmonary infarction in patients with myocardial insufficiency, and later we also wish to record the results of some experiments which deal with the underlying mechanism of jaundice in such cases.

REPORT OF CASES

Case 1 A man with severe myocardial insufficiency, condition unimproved with usual therapy, three days after pulmonary infarction sudden appearance of jaundice and urobilinuria, progressive myocardial failure, death sixteen days after pulmonary infarction.

History A colored man, aged 52 years, with no previous history of syphilis, entered the Johns Hopkins hospital complaining of shortness of breath and swelling of the legs. For one year he had had symptoms of myocardial insufficiency, these symptoms having become considerably worse during the past month.

On examination, he was found to have the physical signs of aortic insufficiency together with the usual evidences of moderately severe congestive heart failure. The liver was enlarged. The sclerae were not icteric. There was no anemia and the white blood count was 10,800 per cmm. The urine contained neither bilirubin nor urobilin.

Course before pulmonary infarction Following the usual forms of therapy the patient's condition remained stationary. He had numerous attacks of paroxysmal dyspnea.

Course following pulmonary infarction On the fourth day after admission, the patient was seized with a pain in the chest, which was followed by tachycardia, increased respiratory distress, paroxysmal coughing and the expectoration of bloody sputum. The temperature rose a degree and one-half above the previous average temperature (99° to 101°F). *Three days following the infarction the sclerae, became jaundiced and urobilin appeared in the urine.* The jaundice deepened, myocardial insufficiency increased and death occurred sixteen days after the development of the infarction.

Necropsy findings The important necropsy findings were syphilitic aortitis extending to the vessels of the aortic arch, scars in the myocardium, cardiac hypertrophy, mural thrombi in all the heart chambers, infarct of the spleen, *embolic thrombi in the pulmonary arteries with multiple infarctions of the lungs, bronchopneumonia, extreme chronic passive congestion of the liver.*

Case 2 A man with severe myocardial insufficiency, temporary improvement, two days after pulmonary infarction appearance of jaundice, bilirubinuria and urobilinuria, hemorrhages into skin, progressive myocardial failure leading to death seven days after pulmonary infarction.

History A white man, aged 37 years, with a previous history of acute rheumatic fever, entered the hospital complaining of shortness of breath and swelling of the legs. He had had symptoms of myocardial insufficiency for six months.

On examination, he was found to have the physical signs of aortic valvular disease, together with the usual signs of severe myocardial insufficiency of the congestive type. There was deep cyanosis but no jaundice. The liver was enlarged. There were frequent attacks of coughing with the expectoration of blood tinged sputa. There was no anemia and the white blood cells were 12,400 per cmm. The urine did not contain any bilirubin.

Course before pulmonary infarction For three days after admission, there was definite improvement in his condition, the edema diminished, the blood tinged sputa disappeared, and cyanosis became less marked.

Course following pulmonary infarction Three days after admission the patient had a chill, a severe attack of coughing and he expectorated a small amount of blood. His respiratory rate increased, the pulse rate became more rapid, and cyanosis deepened, these symptoms were unaccompanied by pain or fever. On the following day a petechial eruption appeared on the chest, abdomen and the upper part of the legs and arms.¹ The severity of myocardial insufficiency in-

¹ The eruption was part of the hemorrhagic phenomena occasionally seen in patients with myocardial insufficiency and particularly in patients with myocardial insufficiency and jaundice. These hemorrhages may be due in part to extensive liver damage.

creased. Two days following the pulmonary infarction the sclerae were yellow and on the next day the skin was definitely jaundiced and bilirubin was found in the urine. The petechial eruption became more extensive and purpuric spots appeared over the back. There was marked chemosis with hemorrhages into the conjunctivae. Jaundice continued to deepen. Blood cultures were negative. The temperature remained normal. Death occurred seven days after infarction.

Necropsy findings The important findings at necropsy were chronic aortic stenosis and insufficiency, marked chronic passive congestion of the viscera, mural thrombi in the right auricular appendix, ascites, multiple old and fresh hemorrhagic infarcts in the lungs, marked chronic passive congestion of the liver with necrosis of the cells in the central portion of the lobules, jaundice.

Case 3 A girl with severe myocardial failure, probable history of pulmonary infarction two days before admission, on admission, slight jaundice and bilirubinuria, temporary improvement with disappearance of bilirubinuria, sudden deepening of jaundice on day following another pulmonary infarction, reappearance of bile in urine, progressive myocardial failure, death three days later.

History A white girl, aged 20 years, with a previous history of acute rheumatic fever, entered the hospital complaining of shortness of breath, palpitation, and edema of the legs. For six years, there had been symptoms of myocardial failure which, following the birth of a child, increased so markedly that she was obliged to enter the hospital. Two days before admission, she had had severe pain in the chest, followed shortly by cough, blood tinged sputum, and an exacerbation of her general symptoms.

On examination, she was found to have the physical signs of mitral, aortic and probably tricuspid valvular disease, associated with the usual evidences of severe congestive heart failure. The liver was enlarged, and there was a slight icteric tinge to the sclerae. There was a moderate secondary anemia (hemoglobin 65 per cent), and the white blood cells were 21,000, per cmm. The urine contained a trace of bilirubin. The stools contained bile.

Course before pulmonary infarction Following the usual forms of therapy, there ensued a period of slight improvement, during which the bile disappeared from the urine.

Course following pulmonary infarction On the twelfth day after admission, she suffered from sudden pain in the chest, the pulse became more rapid, cyanosis deepened, and she developed paroxysms of coughing, productive of bloody sputum. There was no elevation of temperature above the previous level (99° to 101°F). On the following day, there was a striking increase in the degree of jaundice and bilirubin reappeared in the urine. These symptoms were associated with marked increase in the severity of myocardial failure, which progressed up to her death, three days later.

Necropsy findings The important necropsy findings were cardiac hypertrophy, chronic endocarditis of mitral, aortic and tricuspid valves, chronic passive congestion of the lungs, multiple hemorrhagic infarcts of the lungs, bronchopneu-

monia, ascites, fibrinous pleurisy over both lungs, *extreme chronic passive congestion of the liver with necrosis of the central part of the lobules and fatty infiltration of the midzone of the lobules, jaundice*

Case 4 A man with moderately severe myocardial insufficiency and slight jaundice, probable pulmonary infarction two days before admission, temporary improvement, intensification of jaundice three days following pulmonary infarction, appearance of bilirubinuria, hemorrhages into skin, slight fever, death eleven days later, due to progressive myocardial failure

History A white man, aged 39 years, whose previous history was unimportant, entered the hospital on account of shortness of breath and edema of the legs. He had had symptoms of myocardial insufficiency for nine months. Two days before admission all of his symptoms had been exaggerated following the occurrence of pain in the chest that was followed by the expectoration of bloody sputum.

On examination, the striking features were the physical signs of a dilated aorta with aortic insufficiency associated with a moderate degree of heart failure of the congestive type. The sclerae had a yellowish tinge but the urine did not contain bilirubin. There was neither anemia nor fever.

Course before pulmonary infarction Following the usual forms of therapy, the patient's general condition improved for a period of five days. Edema diminished and cyanosis became less marked.

Course following pulmonary infarction Five days after admission the patient was seized with pain in the chest, associated with extreme respiratory distress. The pulse rate increased in frequency, the respirations became more rapid and labored, cyanosis deepened and edema increased. There were frequent attacks of coughing with expectoration of bloody sputum. *Three days after infarction, the jaundice, which had been very faint, became intense, bilirubin appeared in the urine and a few petechial hemorrhages appeared about the knees.* The jaundice was most marked over the upper half of the body and not so conspicuous over the legs.² The temperature rose to 101°F three days after infarction. Death occurred eleven days later.

Necropsy findings The necropsy revealed syphilitic aortitis with involvement of the aortic valves, chronic passive congestion of the viscera, *extensive hemorrhagic infarcts of both lungs, jaundice, marked chronic passive congestion of the liver with necrosis of cells about the center of the lobule and increase in the connective tissue*

² We have observed upon several occasions that the jaundice in patients with myocardial insufficiency is most conspicuous over the upper portions of the body where the edema is less marked. It is most marked in the sclerae and mucous membranes, in the face and arms and over the chest and abdomen but to a less extent over the edematous legs. Occasionally it is seen best upon the abdominal wall, where jaundice is not masked by cyanosis and the tissues are not very edematous. This distribution of jaundice in myocardial insufficiency has also been noted by Meakins (1925).

Case 5 A man with moderately severe myocardial insufficiency, no improvement, two days following pulmonary infarction sudden appearance of jaundice with increase in bilirubin content of blood, appearance of bilirubinuria, progressive myocardial failure, death eleven days after pulmonary infarction

History A white man, aged 45 years, with a previous history of syphilis, entered the hospital complaining of abdominal pain and spells of dizziness. He had had attacks of paroxysmal dyspnoea and substernal pain for ten months. He had had one previous attack of congestive heart failure from which he had recovered.

On examination, there were present the physical signs of syphilitic aortitis with aortic insufficiency, associated with signs of moderately severe myocardial failure. The liver was enlarged. There was no jaundice and the urine was free from bilirubin. There was no anemia and the white blood cells were 8,600 per cmm.

Course before pulmonary infarction The patient's condition remained stationary for seven days. He had irregular fever, numerous attacks of paroxysmal dyspnoea, and substernal pain.

Course following pulmonary infarction Seven days after admission the patient's temperature rose to 104°F, he complained of pain in his chest and expectorated bloody sputum. On this day the amount of bilirubin in the blood serum was two units (van den Bergh), giving the indirect reaction. Two days later the sclerae and skin were visibly jaundiced and bilirubin appeared in the urine. The amount of bilirubin in the blood on this day was four units and the reaction was direct.

On the fifth day following infarction the blood gave the direct reaction and contained five units of bilirubin, and the skin was now deeply jaundiced. He failed progressively and died eleven days after infarction.

Necropsy findings The important necropsy findings were syphilitic aortitis with aortic insufficiency, cardiac enlargement, chronic passive congestion of the viscera, old and fresh pulmonary infarcts, jaundice, terminal pneumonia, chronic passive congestion of the liver with necrosis of the cells about the central vein.

Case 6 A man with severe myocardial failure, slight jaundice and urobilinuria on admission, temporary improvement, on day following pulmonary infarction sudden deepening of jaundice with appearance of bilirubinuria, progressive heart failure leading to death fifteen days after pulmonary infarction.

History A white man, aged 41 years, with a history of recurring attacks of acute rheumatic fever, entered the hospital on account of edema of the legs and a cough without the production of sputum. There had previously been several attacks of myocardial insufficiency, each time with temporary recovery. His present attack was of four months duration.

On examination he was found to have the signs of mitral valvular disease, auricular fibrillation and severe myocardial insufficiency of the congestive type. There was definite jaundice of the skin and sclerae. There was a moderate secondary anemia (hemoglobin 70 per cent), the leucocytes were 7,640 per cmm. There was urobilinuria and moderate retention of nitrogen in the blood.

Course before pulmonary infarction There was a definite improvement in the patient's condition for four days. As cyanosis disappeared, icterus became more distinct.

Course following pulmonary infarction Four days after admission the patient had severe pain in the chest, the respirations became accelerated, the heart rate increased, the pulse deficit became more marked, cyanosis deepened but the temperature did not rise above the previous level (100° to 100.6°F). There was neither cough nor bloody expectoration. *On the following day there was a marked increase in the degree of jaundice, bilirubinuria appeared, and a friction rub was heard over the lower lobe of the right lung.* The myocardial insufficiency increased in severity and the patient died fifteen days later.

Necropsy findings The important necropsy findings were chronic endocarditis of the mitral and tricuspid valves, *marked chronic passive congestion of the liver, infarct in the right median lobe, right hydrothorax.*

Case 7 A man with severe myocardial failure and slight jaundice, temporary improvement, sudden increase of jaundice two days following pulmonary infarction, appearance of bilirubinuria, hematemesis and melena, progressive myocardial failure, death six days after pulmonary infarction.

History A white man, aged 37 years, with a history of acute rheumatic fever, entered the hospital on account of swelling of the extremities and breathlessness. He had had symptoms of myocardial insufficiency for four months.

On examination he was found to have the physical signs of aortic and mitral valvular disease, associated with rather severe myocardial insufficiency of the congestive type. The sclerae and skin had a yellowish tinge, the urine did not contain bilirubin. There was no anemia and the white blood cells were 10,400 per cmm.

Course before pulmonary infarction Following the usual forms of therapy there was a definite improvement in the patient's condition. The edema diminished, the liver became smaller and cyanosis was less marked.

Course following pulmonary infarction Four days after admission, the patient suddenly became more dyspneic, the pulse became more rapid, cyanosis deepened and the signs of congestion increased. There was no fever, pain, cough nor bloody expectoration. *Within two days the jaundice deepened, urobilinuria and bilirubinuria appeared.* The patient had a hematemesis and the stools contained occult blood. Death occurred six days following pulmonary infarction.

Necropsy findings The necropsy revealed chronic mitral and aortic stenosis, cardiac hypertrophy, chronic passive congestion of the viscera, mural thrombi in the right auricular appendage, *multiple hemorrhagic infarcts of the lungs, jaundice, chronic passive congestion of the liver with necrosis of the cells about efferent vein of the lobule, multiple superficial gastric and duodenal ulcers, diphtheritic colitis.*

Case 8 A man with severe myocardial failure and slight jaundice, temporary improvement, two days following pulmonary infarction deepening of jaundice and

appearance of bilirubinuria, progressive myocardial failure Death nine days following pulmonary infarction

History A colored man, aged 41 years, with a history of acute rheumatic fever, entered the hospital on account of swelling of the extremities. He had had symptoms of myocardial insufficiency for three months and for one month they had been severe.

On examination, he was found to have the signs of mitral valvular disease and moderately severe myocardial insufficiency. The sclerae had a yellowish tinge, there was no bilirubinuria. There was no anemia and the white blood cells were 8,400 per cmm.

Course before pulmonary infarction Following the usual forms of therapy there was a definite improvement in the patient's condition for a period of four days.

Course following pulmonary infarction Four days after admission the patient had pain in the chest which was followed by tachycardia, increased respiratory rate, cough and bloody sputum. The temperature did not rise above the former level (99° to 100°F) until three days after the infarction took place when the temperature was 102°F. The white blood count was 8,040 per cmm. The symptoms of myocardial insufficiency increased, *four days after infarction the jaundice deepened and bilirubin appeared in the urine.* The patient failed rapidly and died nine days after pulmonary infarction.

Necropsy findings The necropsy revealed chronic mitral endocarditis, adhesive pericarditis, cardiac enlargement, scars in the myocardium, *multiple hemorrhagic infarcts of the lungs, jaundice marked chronic passive congestion of liver with necrosis of central and mid zones of the lobules, bronchopneumonia*

Case 9 A man with severe myocardial failure, sudden appearance of jaundice two days following pulmonary infarction, temporary improvement for sixteen days, deepening of jaundice and appearance of bilirubinuria following second pulmonary infarction, hematemesis, death due to progressive myocardial failure seven days after second pulmonary infarction.

History A white man, aged 59 years, whose past history was unimportant, entered the hospital on account of swelling of the abdomen. He had had one previous attack of myocardial failure five months before admission. He had had symptoms of moderately severe myocardial insufficiency for two months.

On examination, the patient was found to have signs of aortic valvular disease, with a dilated aortic arch associated with the usual evidences of myocardial insufficiency of the congestive type. There was no jaundice. The liver was enlarged. The urine did not contain any bilirubin and there was no anemia. The white blood cells were 5,880 per cmm.

Course before pulmonary infarction Following the usual methods of therapy the patient's condition improved for a period of two days.

Course following pulmonary infarction Two days after admission the patient began to have attacks of coughing and expectorated bloody sputum. The respiratory and heart rates increased, but there was no fever. Two days later, *there*

was definite jaundice of the sclerae and skin and urobilinuria but no bilirubinuria Following this attack his general condition improved for a period of sixteen days when another pulmonary infarct occurred. This was followed by an increase in myocardial insufficiency, jaundice became more intense and bilirubin appeared in the urine. The patient had a hematemesis two days before death and died seven days after the second pulmonary infarction.

Necropsy findings The important necropsy findings were syphilitic aortitis, aortic insufficiency, aneurysm of the arch of the aorta, chronic adhesive pericarditis over the aneurysm of the ascending arch of the aorta, chronic passive congestion of the viscera, multiple infarcts in the right lung, hemorrhagic erosions of the mucosa at the cardiac end of the stomach, jaundice, chronic passive congestion of the liver with necrosis of the cells about the central vein.

Case 10 A man with severe myocardial insufficiency, slight jaundice on admission, temporary improvement, two days following pulmonary infarction increase in intensity of jaundice, appearance of bilirubinuria, death twelve days later due to progressive myocardial failure.

History A colored man, aged 45 years, was admitted to the hospital on account of swelling of the extremities. He had had symptoms of progressive myocardial failure for one year and these symptoms had all become more severe recently.

On examination, he was found to have an enlarged heart without evidences of valvular disease, moderate arteriosclerosis and the usual signs of myocardial insufficiency of the congestive type. There was a slight yellowish tint of the sclerae. The urine did not contain bilirubin. The liver was enlarged. The amount of bilirubin in the blood at this time was 1.5 units (indirect).

Course before pulmonary infarction Following the usual forms of therapy, his general condition seemed to improve. His liver remained large, however, jaundice of the sclerae deepened in degree, the amount of bilirubin in the blood rose to 3 units (indirect) and urobilin appeared in the urine.

Course following pulmonary infarction Eight days after admission he became more dyspneic, he had paroxysmal attacks of coughing with the expectoration of bloody sputum. The symptoms of myocardial insufficiency increased and auricular fibrillation appeared. Three days later, jaundice became more marked, the amount of bilirubin in the blood increased to five units (direct and bilirubin appeared in the urine). The myocardial insufficiency and jaundice continued to increase in degree, a friction rub appeared over the lung. The temperature was not elevated above 101°F. The leucocyte count was 7,800. Two days before death there were 10 units (direct) of bilirubin in the blood. Death occurred twelve days after pulmonary infarction.

Necropsy was not obtained.

ANALYSIS OF CLINICAL CASES

a Clinical picture before pulmonary infarction Of the ten cases studied in this group, five had syphilitic, four rheumatic and one ar-

teriosclerotic heart disease. One-half of these patients had had previous attacks of myocardial insufficiency varying in duration from two months to one year before the illness that brought them to the hospital. All had had symptoms of heart failure from one to three months before coming under observation. Four of the ten patients gave a history of having had symptoms that might be interpreted as being due to previous pulmonary infarctions. On examination, seven of the ten patients had severe heart failure, while the other three were moderately ill. Six of the patients had slight jaundice of the sclerae and skin, while four showed no evidence of jaundice whatever. The liver was enlarged in eight cases and not palpable in the other two. Bilirubin was present in the urine in only two cases. The leucocyte count was normal in all of the cases except one in which there was a leucocytosis of 21,000 per cmm. Following the usual forms of therapy six of the patients improved, three remained stationary and one patient became progressively worse.

b Clinical picture following pulmonary infarction Following the development of pulmonary infarction, the entire clinical picture changed, suddenly and often without warning. The characteristic features and course were 1, increased severity of the manifestations of myocardial failure, 2, the appearance of jaundice, 3, the symptoms dependent on the local process in the lungs, and 4, the appearance of hemorrhagic phenomena. The first one was constant, aside from the appearance of jaundice, the most characteristic feature was the sudden exacerbation of the symptoms of myocardial insufficiency, rapidly progressive, and leading to the death of the patient within three to fifteen days. Marked cyanosis was present in every case. In one patient transient auricular fibrillation developed.

The symptoms usually considered characteristic of pulmonary infarction were not always striking, and were, in fact, sometimes absent. In practically all cases, there was increase in the respiratory rate and also in the severity of dyspnea, but ordinarily, these symptoms alone are not necessarily an indication of the development of pulmonary infarction. Thoracic pain was present in six cases, cough and bloody expectoration in eight. In only four instances did a friction rub appear over the affected lung, in these cases it was heard within two to five days after infarction occurred. Fever was present in only four

cases, and was not associated with marked increase in the number of leucocytes

The most striking and conspicuous manifestation of pulmonary infarction in these cases was the sudden appearance of jaundice, or, if jaundice was already present, its sudden deepening. This symptom occurred within one to four days, usually on the second or third after infarction. About the same time, bilirubin was found in the urine of all but one of the cases, and in this exceptional instance, there was a large amount of urobilin in the urine.

In two cases hemorrhages occurred into the skin, in two cases hematemases occurred, and melena and hematuria developed, each in one instance.

The important necropsy findings in nine of our cases were pulmonary infarctions, usually multiple and extensive chronic passive congestion of the liver with necrosis of the cells about the central veins. The bile passages were free and patent in all of the cases. The source for the pulmonary emboli was the right auricular appendage in five cases and in the other four cases the source for the emboli could not be determined with certainty.

CLINICAL PICTURE

Clinical picture With these data, one may construct the following picture. The patient suffers from fairly severe heart failure, usually of considerable duration. At first he may show slight jaundice, visible jaundice may however be entirely absent. Under treatment, definite improvement may take place. Suddenly, a distinct change in the clinical picture occurs, dependent on the development of pulmonary infarction. Dyspnea becomes more intense, cyanosis deepens, myocardial failure increases and progresses rapidly. Usually, on the second or third day after the occurrence of pulmonary infarction, jaundice suddenly makes its appearance, or it becomes markedly intensified if it has been present previously. At the same time, bilirubin and urobilin are found in the urine. Hemorrhagic phenomena occasionally develop. These symptoms increase, advance steadily, and within a relatively short time, death ensues. It is important to bear in mind the fact that the entire course of events may take place in the absence of characteristic signs of pulmonary infarction. *The*

sudden appearance of jaundice or the sudden increase in the severity of myocardial insufficiency, should make one suspect that pulmonary infarction has occurred, even when outspoken evidences of pulmonary infarction cannot be obtained

Undoubtedly, the above description applies only to a certain group of patients. There are certainly instances of pulmonary infarction which bring about lesser degrees of jaundice, perhaps detectable only by use of a method such as the van den Bergh test, and it is altogether probable that in these cases, the clinical course is not so stormy as we have pictured it, and that recovery frequently takes place.³ Indeed, in our own cases, there were several instances in which the slight jaundice that was first seen was probably due to previous infarctions (possibly multiple), and recovery, at least temporary, might have ensued, had not further infarction taken place, this time leading to irreparable damage and death.

SUMMARY AND CONCLUSIONS

We have reported 10 cases of myocardial insufficiency in which the sudden appearance of visible jaundice, or the sudden increase in jaundice that was already present seemed directly dependent on the development of pulmonary infarction. The jaundice was due, in all instances, to bilirubinemia. In all these cases, the clinical course was very similar, for death, due to progressive myocardial failure, occurred within a relatively short time.

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³ Since this paper was written, we have observed four more patients in whom jaundice developed following pulmonary infarction. Three of the patients recovered and one died. We should like to emphasize the fact, therefore, that pulmonary infarction occurring in patients with myocardial insufficiency and followed by jaundice does not lead to a fatal outcome in all instances.

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JAUNDICE FOLLOWING PULMONARY INFARCTION IN PATIENTS WITH MYOCARDIAL INSUFFICIENCY

II AN EXPERIMENTAL STUDY

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(Received for publication January 30, 1926)

INTRODUCTION

In a preceding paper (Keefer and Resnik (1926)) we described ten patients suffering from myocardial insufficiency, in whom jaundice appeared for the first time or increased considerably if already present following the development of pulmonary infarction.

It is apparent that pulmonary infarction in a patient suffering from heart failure involves more than the mere extravasation of blood into the lungs. The experimental work of Binger, Brow and Branch (1924) shows that the chief functional disturbance which occurs as a result of pulmonary embolism is the development of anoxic anoxemia, a state in which the arterial blood contains less than its normal quota of oxygen. Undoubtedly, anoxemia of a similar kind developed in the patients whose histories we have reported. While it is true that in the experiments of Binger, Brow and Branch it was only after multiple embolisms had taken place that anoxemia appeared, and that in some of our patients on the contrary only one infarct occurred, it must be remembered that the circumstances differ in the two instances. In the experiments of Binger, Brow and Branch, the animals and the lungs were normal. A considerable amount of mechanical obstruction of the pulmonary circulation must take place before the functional impairment of the lungs is sufficiently great to produce anoxic anoxemia. On the other hand, it is altogether reasonable to assume that, in patients such as we have described, in whom there is already functional impairment of the lungs, due to chronic passive congestion, still further

diminution in the amount of effective pulmonary tissue may bring on anoxic anoxemia, or exaggerate the degree of anoxemia already present

There is another factor of prime importance. All the patients in whom jaundice appeared following pulmonary infarction died within a relatively short time of progressive myocardial failure. There is evidence to show that anoxemia depresses myocardial function, particularly when the myocardium is already damaged (Resnik, 1925), so that the effect of pulmonary infarction in patients with myocardial failure is not only to cause anoxic anoxemia to appear, but also to increase the degree of stagnant anoxemia, which is dependent on the development of diminished circulatory minute volume.

Finally, the condition of the liver remains to be considered. Since all our patients were suffering from severe and long standing myocardial insufficiency, it is probable that at the time when pulmonary infarction developed there must have been present changes in the liver characteristic of chronic passive congestion. Since according to contemporary theories of jaundice (McNee, 1922), (Rich, 1925), it is the function of the hepatic cells to excrete the bile pigment that is brought to them by the blood stream, the functional efficiency of these cells must be kept clearly in mind.

In pulmonary infarction in patients with myocardial failure, we must deal, then not only with extravasation of blood into the pulmonary tissues, but with the functional disturbances that such a lesion may bring about. To state the case more directly *It is possible that anoxemia, which is caused by pulmonary and circulatory impairment, resulting from pulmonary infarction, may depress the excretory function of already damaged liver cells to such an extent that jaundice appears.* The experiments now to be described had as their object a study of the effect of anoxemia on the damaged liver.

EXPERIMENTAL METHODS

To produce a liver lesion that would simulate the condition of chronic passive congestion, we had recourse to the use of carbontetrachloride. When this substance is given by mouth, a central lesion of the liver lobule occurs (Lamson, Gardner, Gustafson, Maire, McLean and Wells (1923)), (Gardner, Grove, Gustafson, Maire, Thompson, Wells and Lamson, (1925)). The liver injury reaches its maximum

within 24 to 48 hours, and healing begins three or four days after administration

Dogs were given carbontetrachloride by stomach tube, in doses of 4 cc per kilo of body weight. Twenty-four hours later, they were anesthetized by the subcutaneous injection of morphine, approximately 6 mg per kilo, followed by the administration of urethane, 1 gm per kilo, given in watery solution by stomach tube

Through a Y-shaped tracheal cannula, a catheter was inserted down to the bifurcation of the trachea, the catheter being connected with a tank of nitrogen. Anoxemia, of any desired degree, was produced by varying the amount of nitrogen flowing into the lungs. The degree of anoxic anoxemia was determined by analysis of samples of blood taken from the femoral artery. Analyses of the oxygen content and capacity of the blood were made by the methods of van Slyke. On a few occasions, similar analyses of the venous blood were made, in order to ascertain the degree of stagnant anoxemia. Samples of blood for the determination of the bilirubin content were taken usually from the femoral vein, occasionally from the femoral artery. The blood was drawn into tubes containing a few crystals of sodium oxalate, the amount of bilirubin being estimated by the modification of the van den Bergh test recently described by McNee and Keefer (1925). When the urine was tested in order to find whether bile pigment was present, the Salkowski modification of the Huppert test was employed. In several experiments, the presence of hematin in the blood was sought, by spectroscopic analysis of the plasma, using the Leitz spectroscope attachment for the microscope.

The general procedure in the experiments was to estimate the bilirubin content of the blood at intervals, while the animal was breathing air, usually with an added quota of oxygen¹. After several hours, anoxemia was induced, and bilirubin determinations of the blood were made again at intervals. Since the exact procedure differed in individual experiments, the details are given in the protocols. On completion of an experiment, pieces of liver were removed for histological examination, and other observations were made which will be recorded in the protocols.

¹ It will be understood that whenever the bilirubin content of the blood is mentioned, it is actually to the blood plasma or serum that the reference is made.

RESULTS OF EXPERIMENTS

Dog 1 Weight 11 pounds This animal was used as a control In order to test the effect of anoxemia on the normal liver carbontetrachloride was not given At the beginning of the experiment, 35 cc of the animal's own blood was injected intraperitoneally, considering the size of the dog, this was the equivalent of a fairly large extravasation of blood This was done because in some of the other experiments, there was extravasation of a certain amount of blood into the peritoneal cavity due to operative procedure, although it was improbable

TABLE 1

Dog 1 Effect of anoxemia on the bilirubin content of blood of an animal with a normal liver

Time	Oxygen saturation of arterial blood	Bilirubin content of blood plasma	Hematin in blood plasma	Remarks
	<i>per cent</i>	<i>units</i>		
9 45 a.m.	95*	0	0	
11 15 a.m.	56			Flow of nitrogen to lungs started
11 35 a.m.	56	0	0	
1 55 p.m.	56	0	0	
3 15 p.m.	56	0	0	
5 00 p.m.	56	0	0	
5 01 p.m.	<56			Flow of nitrogen increased
6 00 p.m.	56	0	0	
6 01 p.m.	56			Flow of nitrogen further increased
6 30 p.m.	56	0	0	
6 35 p.m.				Animal died

* This figure is approximate, the arterial blood was at least 95 per cent saturated with oxygen, since the breathing mixture was enriched with oxygen

that, during the relatively short duration of a single experiment, the presence of blood in the peritoneal cavity could have had any effect on the bilirubin content of the blood

The results of the experiment are summarized in table 1

Autopsy The bile passages were patent The urinary bladder contained 80 cc urine, which was free of bile pigment Blood was present in the peritoneal cavity, approximately similar in amount to that injected *Histological examination of the liver showed slight but definite central necrosis of the liver lobules* We shall return later to a discussion of the significance of this finding

Comment Extremely severe anoxemia, of over seven hours duration, had no effect in increasing the bilirubin content of the blood of this animal, whose liver, at the beginning of the experiment was normal. There was moreover no evidence that the presence of free blood in the peritoneal cavity caused the appearance of hematin in the blood during the period of the observations.

Dog 2 Weight 25 pounds, 4 cc carbontetrachloride administered 24 hours previously

TABLE 2

Dog 2 Effect of anoxemia on the bilirubin content of the blood of an animal with injured liver

Time	Oxygen saturation of arterial blood	Bilirubin content of blood plasma	Remarks
	<i>per cent</i>	<i>units</i>	
10 10 a.m.	92	0	Hemolyzed blood in plasma of every sample of blood
1 10 p.m.	92	0	
3 00 p.m.	92		
3 10 p.m.	92	0	
4 10 p.m.	93	0	
4 50 p.m.	93	0	
5 10 p.m.	72		Arterial anoxemia*
5 55 p.m.	72	0	
7 35 p.m.	72	Trace	
8 46 p.m.	<72†	0 75	
9 45 p.m.	72	0 75	
10 45 p.m.	72	0 75	
11 35 p.m.	72	0 75	Immediately after death

* Actually, the flow of nitrogen was started at 2 00 p.m. Analyses of the arterial blood at 3 00 p.m. and at 4 00 p.m. showed, however, that the arterial blood was normally saturated. This was undoubtedly due to the fact that respiration was stimulated sufficiently to compensate for the slightly increased percentage of nitrogen in the inspired gas. At 5 10 p.m. the percentage of nitrogen in the breathing mixture was increased, analysis at this time showing that the arterial blood was 72 per cent saturated. Anoxemia must be considered, then, to have begun at this time.

† From 8 00 p.m. on, there was present not only anoxic anoxemia, but also stagnant anoxemia, the extent of which was not determined. We make this statement, because the animal was in poor condition. In order to prolong the experiment as long as possible, the flow of nitrogen was discontinued at 8 00 p.m. and at 10.30 p.m. for about 10 minutes each time. During this time, a rich mixture of oxygen was given. In spite of the fact however that the arterial blood was rendered bright pink, the mucous membrane of the tongue remained distinctly cyanotic, indicating that the blood flow through the tissues was impaired. The degree of anoxemia was really greater, then, than would appear from the figure in the table.

Autopsy The bile passages were patent. The urinary bladder held 120 cc urine which contained a trace of bile pigment (the exact amount was not determined). The lungs were normal. Histological examination of liver tissue show marked central necrosis of the liver lobule.

Comment In this animal in which the liver had been damaged by the administration of carbontetrachloride, there was no bilirubin in the blood over a period of seven hours when the arterial blood was normally saturated with oxygen. Two and a half hours after inducing a fairly severe anoxemia, there was a trace of bilirubin in the blood, the amount increasing to 0.75 unit an hour later. Thereafter there was no increase. The anoxemia which was present was not only of the anoxic type, but also of the stagnant type.

On centrifuging each specimen of blood, it was found that there was constantly present a slight amount of hemolyzed blood in the supernatant plasma. That this was not due to oxalate is shown by the fact that a similar finding was observed when the blood was allowed to clot.² *Although bilirubinemia might have occurred owing to injury of the liver and probably an increase in the formation of bile pigment due to the presence of free hemoglobin in the circulating blood, bilirubinemia did not appear until anoxemia was induced.*

Dog 3 Weight 41 pounds, 4 cc per kilo of carbontetrachloride given 24 hours previously. Kidney pedicles tied. In the experiment just described (dog 2), there exists the possibility, even though unlikely, that the increase of bilirubin in the blood was due partly to depression of kidney function as the result of anoxemia. Because of this possibility, the entire pedicle of each kidney was securely ligated.³

Autopsy The bile passages were patent. The renal artery, vein, and the ureter were securely ligated on both sides. There was about 25 to 30 cc of free blood in the peritoneal cavity, due to the operative procedure. The lungs were normal. Histological examination of the liver showed central necrosis of the liver lobules.

Comment During the control period, when the arterial blood was

² Lamson, Gardener et al (1923) have also found intramuscular hemolysis, although apparently less frequently than we have.

³ This was done, rather than removal of the kidneys, in order to avoid, so far as possible, surgical shock.

TABLE 3

Dog 3 Effect of anoxemia on the bilirubin content of the blood of an animal with injured liver

Time	Oxygen saturation of arterial blood	Bilirubin content of blood plasma	Remarks
	<i>per cent</i>	<i>units</i>	
10 25 a.m.	95?*	Faint trace?	Hemolyzed blood in plasma of every sample of blood
1 35 p.m.	95	Faint trace?	
2 00 p.m.	57		Flow of nitrogen started
2 35 p.m.	57	Definite trace	
3 05 p.m.	57	1 1	
3 45 p.m.	57	1 5	Sample taken immediately after death

* Analysis not done. Dog was breathing air enriched with oxygen.

TABLE 4

Dog 4 Effect of anoxemia on bilirubin content of the blood of an animal with injured liver

Time	Oxygen saturation of arterial blood	Bilirubin content of blood plasma	Remarks
	<i>per cent</i>	<i>units</i>	
10 30 a.m.	95?*	Faint trace	Condition poor Tongue cyanotic in spite of high percentage of oxygen in breathing mixture
1 30 p.m.	95	1 56	
1 50 p.m.	95		Condition of animal poor Oxygen saturation of the femoral vein 35 7 per cent
2 30 p.m.	95	1 50	
3 38 p.m.	95	1 56	
4 30 p.m.	87 4		Added flow of oxygen discontinued Animal breathing air Respirations shallow
4 35 p.m.	87 4	1 50	
5 35 p.m.	87 4	1 60	
6 45 p.m.	87 4		
7 45 p.m.	87 4	1 75	
8 45 p.m.	87 4	1 88	
9 45 p.m.	87 4	2 8	
11 35 p.m.	87 4	1 56	Sample taken immediately after death

* Oxygen content was not done. Animal was inspiring an oxygen rich mixture of gas The arterial blood was bright pink and was probably 95 per cent saturated with oxygen

normally saturated with oxygen, there was a questionable trace of bilirubin in the blood. One-half hour after severe anoxemia was induced, there was a definite increase in the amount of bilirubin of the blood, the amount reaching 1.5 units at the time of death of the animal, about two hours later.

Dog 4 Weight 12 pounds, carbontetrachloride, 4 cc per kilo, given by stomach tube 24 hours previously. Kidney pedicles tied.

Autopsy The bile passages were patent. There was free blood, 15 to 20 cc, in the peritoneal cavity. The entire kidney pedicle was securely ligated on both sides. Histological examination of the liver showed extensive central necrosis of the liver lobule.

Comment Because of profound operative shock, there was present throughout the entire experiment a considerable degree of stagnant anoxemia, as evidenced by the low oxygen saturation of the venous blood (see table 4, at 1.50 p.m.) while at the same time the arterial blood was normally saturated with oxygen. This we believe was responsible for the increase in the bilirubin content of the blood from a trace to 1.56 units three hours later. At 4.30 p.m., the added supply of oxygen to the inspired air was discontinued. The respirations then became slow and shallow, probably because arterial oxygen saturation diminished to 87.4 per cent. In other words, there was superimposed on stagnant anoxemia, an anoxic anoxemia, although of relatively mild degree. The bilirubin content of the blood then began to increase steadily, reaching its maximum five hours later, the bilirubin concentration of the blood was 2.8 units at this time. About two hours later the animal died. *A sample of blood taken from the heart immediately after death showed that the bilirubin concentration had decreased to 1.56 units.*

Every sample of blood was found to contain free hemoglobin in the plasma. At no time, however, was hematin detected in the plasma by spectroscopic examination.

Dog 5 Weight 11 pounds, carbontetrachloride, 4 cc per kilo, administered 24 hours previously. In order to avoid the operative shock consequent on ligating the kidney pedicles, a cannula was inserted in the urinary bladder. In this way, changes in the bile pigment content of the urine could be detected at intervals during the experiment.

Autopsy The bile passages were patent. There was no blood in

the peritoneal cavity Near the roots of the lungs there were a few small patches of edema and atelectasis Histological examination of the liver showed central necrosis of the liver lobule

Comment During the control period, when the arterial blood was normally saturated with oxygen, the faint trace of bilirubin in the blood, which was present on the first examination, disappeared One hour after the beginning of anoxemia, bilirubin reappeared in the blood, increasing to 0.8 unit shortly before the death of the animal, about six hours after anoxemia was produced

TABLE 5

Dog 5 Effect of anoxemia on the bilirubin content of the blood of an animal with injured liver

Time	Oxygen saturation of arterial blood	Bilirubin content of blood plasma	Bilirubin content of urine	Remarks
	<i>per cent</i>	<i>units</i>		
10 30 a.m.	95 ²	Faint trace?	0	Content not determined Breathing an oxygen rich mixture. Condition of animal excellent
12 30 p.m.	95	Faint trace?	0	
2 35 p.m.	95	0	0	
4 30 p.m.	95	0		
4 31 p.m.	66			Flow of nitrogen to lungs started
5 30 p.m.	66	Faint trace	0	
6 20 p.m.	66	Definite trace		
8 00 p.m.	66	0.6	Faint trace	
8 45 p.m.	66	0.6		
9 45 p.m.	66	0.75	Faint trace	
10 50 p.m.	66	0.8		
				Sample taken one-half hour before death

There was no intravascular hemolysis in this animal

Dog 6 Weight 19 pounds, carbontetrachloride was not given to this animal In all previous experiments, in which liver injury had been caused by the administration of carbontetrachloride, there was an increase in the bilirubin content of the blood within a relatively short time after anoxemia was instituted The increase of bilirubin in the blood may, it appeared, have been due to increased formation of bilirubin during periods of anoxemia. The damaged liver may, under

ordinary circumstances, have been able to excrete the amount of bilirubin which was normally formed, but during anoxemia the production may have been excessive so that the liver was taxed beyond its capacity, with the result that bilirubin began to accumulate in the blood

The cystic duct was ligated and a cannula was inserted in the common bile duct and connected with a rubber tube leading out through the abdominal wound. The kidneys were not removed or isolated, but the urine in the bladder at the end of the experiment was entirely free of bile pigment. Consequently, by determinations of the bilirubin content of the blood and the bile, we obtained information concerning the amount of bilirubin which was being formed during a given period of time. There was no intravascular hemolysis, nor was there any blood in the peritoneal cavity at autopsy. Abnormal blood destruction was in this case then not the cause of increased bile pigment formation.

Although it has been shown by Rous, Brown and McMaster (20) that erroneous impressions of the output of bile pigment may be gained in bile-fistula experiments, when the periods of collection are of short duration, we were nevertheless obliged to use short collection periods, first, because of the nature of the experiment, and, second, because in our other experiments, the changes in bilirubin content in the blood had occurred within a relatively short period of time after anoxemia was produced. The collections of bile were made in periods of two and three hours. The amount in a single period was made up to a total of 100 cc. with distilled water and the quantity of bilirubin was then determined by the van den Bergh test. The number of units found in the diluted specimen was divided by the number of hours during which the collection was made, giving the number of units of bilirubin per hour. The rubber tube leading to the container in which the bile was collected was not emptied at the end of each collection period. It is possible, therefore, that an error was introduced through the circumstance that the rubber tube may have contained varying amounts of bile at different times. This error, if it existed, must, however, have been slight.

Autopsy The cystic duct was securely ligated and the passage between the common duct and the cannula was patent. There was no

bile pigment in the urine of the urinary bladder There was practically no free blood in the peritoneal cavity *Histological examination of the liver showed slight but definite central necrosis of the liver lobules*

Comment Since the blood and urine were entirely free of bilirubin, the bile which was collected from the common duct must have contained the total output of this pigment We do not wish to stress the actual amounts of bilirubin that were excreted⁴ But the presence of

TABLE 6
Dog 6 Effect of anoxemia on the rate of formation of bilirubin

Period	Time	Duration of collection	Oxygen saturation of arterial blood	Output of bile	Output of bilirubin	Bilirubin content of blood plasma	Remarks
		hours	per cent	cc per hour	units per hour	units	
1	11 40 a.m. to 2 40 p.m.	3	96	4 5	18 7	0	Slight operative shock. Venous blood was 56 per cent saturated with oxygen
2	2 40 p.m. to 5 40 p.m.	3	73	4 06	11 3	0	Condition of animal improving
3	5 40 p.m. to 7 40 p.m.	2	73	3 75	17 0	0	Animal in excellent condition. Required morphia grain $\frac{1}{4}$
4	4 40 p.m. to 9 35 p.m.	2	?	3 15	19 0	0	No gas analysis made. This is a probable figure
5	9 35 p.m. to 11 30 p.m.	2	?	1 5	7 0	0	No gas analysis made. This is a probable figure

⁴McMaster, Broun, and Rous (1923) have found that, in dogs with normal hemoglobin content, the maximum output of bilirubin was 11 mg per kilo of dog in 24 hours If our own figures are translated into corresponding terms, they are for the respective periods 26 4, 16 0, 23 8, 26 5 and 9 8 mg per kilo in 24 hours. Although the conditions differed in the two sets of experiments, we are still unable adequately to account for the greater yield which we obtained Because of this discrepancy, we have hesitated to report this experiment, and to draw conclusions from it which may not be justified, namely that anoxemia may depress the formation of bile pigment. Unfortunately, owing to certain circumstances, we were unable to repeat the experiment, and we have described it because it does seem to explain why the bilirubin content of the blood diminished in experiment 4 even though there was every reason to believe that the liver injury was increasing

anoxemia did not serve to increase the formation of bilirubin. Indeed, the amount of bilirubin obtained during the periods of anoxemia was always less than that obtained when the blood was normally saturated with oxygen. The diminution in bilirubin output was particularly striking in Period 5 when the output fell to about one-third the amount of the preceding period.

SUMMARY AND DISCUSSION OF EXPERIMENTS ON THE EFFECT OF ANOXEMIA

We may now bring together the various findings that have been observed in these experiments. It has been demonstrated that when the liver of a dog has been injured by the administration of carbon-tetrachloride, anoxemia, whether of the anoxic or of the stagnant type, is followed by definite and sometimes striking increase in the bilirubin content of the blood. It has also been shown that severe anoxemia, of several hours duration, fails to bring about this result if the liver has not been previously injured. Finally, it has been shown that the accumulation of bilirubin in the blood is not due to depression of renal function, or to increased formation of bilirubin. We draw the conclusion therefore that in our experiments anoxemia affected the liver in such a way that bilirubinemia appeared, or if it was already present, was exaggerated. The evidence that we possess at the present time indicates that the sole function of the hepatic cell, so far as bilirubin is concerned, is to excrete this pigment. We can, therefore, go further and state that the mechanism by which anoxemia causes bilirubinemia to develop under the conditions of our experiments serves to impair the capacity of the hepatic cell to excrete bilirubin.

There are two observations that need further comment. In the experiment on dog 4 (table 4), the bilirubin content of the blood reached a maximum of 2.8 units during anoxemia. Two hours later, immediately after the animal expired, the bilirubin concentration of the blood had diminished to 1.56 units. The decrease could hardly have been due to improvement in the excretory ability of the liver cells, since the degree of anoxemia was more severe than it had been when the bilirubin content of the blood was 2.8 units. If the function of the liver were the only factor concerned the amount of bilirubin in the blood should have continued to increase. The solution of this

difficulty is apparently supplied by the results obtained in the experiments on dog 6 (table 6) Here it was found in period 5 that after severe anoxemia the formation of bilirubin was sharply diminished Whether the decreased output of bilirubin during this period was due to diminished breakdown of blood pigment, an accepted source of bile pigment, or whether it was due to functional impairment of the agency which transforms blood pigment into bile pigment, we do not know At any rate, anoxemia seems to bring about two mutually antagonistic effects 1, depression of the ability of the liver cell to *excrete* bilirubin, this factor tending to increase the bilirubin content of the blood, 2, depression of bilirubin *formation* (of significance, probably, only in the late stages of anoxemia), tending to decrease the bilirubin content of the blood The result depends on the balance of these effects, and the explanation for the diminution of the amount of bilirubin in the blood in dog 4 lies, undoubtedly, in the fact that the second effect predominated at the end of the experiment.

Another observation of considerable interest was that concerning the influence of anoxemia on the normal liver It will be recalled that the experiments on dogs 1 and 6 were made without the administration of carbontetrachloride, so that the livers of these animals were presumably normal at the beginning of the experiments Yet in both instances, histological examination of liver tissue removed immediately after death showed slight central necrosis This finding probably throws light on the origin of the liver lesion which results from chronic passive congestion Anoxemia of the stagnant type, while it is not the only functional disturbance that occurs as a result of myocardial insufficiency, is a constant one It seems probable, therefore, that *anoxemia may not only impair the function of the already damaged liver, but it may actually be responsible, at least in part, for the damage*

Finally, it may be mentioned that in none of the experiments was hematin found in the plasma although in several there was either intravascular hemolysis or extravasation of blood into the peritoneal cavity, conditions under which hematinemia has previously been found before (Schottmüller 1914)

DISCUSSION

It remains to discuss the clinical bearing of the foregoing observations, particularly with reference to their relation to jaundice in myocardial failure. This may best be done, perhaps, by enumerating the various factors which may enter into the causation of jaundice under these circumstances.

1 Impairment of liver function We refer specifically to the function of the hepatic cell to excrete bilirubin. Experimentally, it has been shown that the degree of bilirubinemia produced by drugs such as carbontetrachloride (Lamson, et al., 1923), which cause a diffuse lesion in the liver is generally proportional to the severity of the liver injury. Clinically, too, we have found that, in general, the same relationship is true. Patients suffering from myocardial failure who show latent or outspoken jaundice are practically always those whose myocardial failure is more or less severe and of long duration. Probably in every case of myocardial failure in which jaundice occurs, one may find extensive anatomical changes in the liver. We believe, therefore, that one of the most important and constant factors in the production of "cardiac" jaundice is the functional impairment of the liver cells, the final result, however, being modified by other influences. It has been pointed out (Fishberg, 1923), that there are patients with intense congestion of the liver, particularly in cases of heart failure of recent origin, in whom the bilirubin content of the blood is not much increased, and, on the other hand, there are patients with high blood bilirubin values in the blood whose livers are not large enough to be palpable. It must be emphasized, however, that the size of the liver does not necessarily give a correct idea of the anatomical changes in the liver or of its functional impairment. It is particularly in the recent cases of myocardial insufficiency with greatly swollen livers that one may find least liver damage on histological examination. Conversely, one may find the most intense central necrosis of the lobules of livers which, because of severe and long standing congestion and consequent necrosis and collapse of the lobules, have become even smaller than normal in size.

2 Anoxemia We have seen that, in the experiments, anoxemia may depress the function of livers already damaged to such an extent

that the bilirubin content of the blood increases strikingly. We have also pointed out that pulmonary infarction may produce anoxemia in patients with severe myocardial insufficiency. It is probably through the effect of anoxemia on liver function that jaundice develops, or increases in patients such as those whose histories we have given in detail in an earlier paper. Anoxemia probably depresses liver function in any circumstance, and we have obtained evidence to show that it may actually be responsible, in part at least, for liver damage in chronic passive congestion. Anoxemia of short duration, even if severe, fails however of itself to cause a sufficiently profound depression of liver function to permit the development of jaundice. There must be, in addition to anoxemia, a certain degree of functional impairment of the liver, in order that jaundice may follow.

3 *Increased destruction of red blood cells* Since Resnik and Rich (1926) were unable to obtain conclusive evidence that local extravasations of blood in patients with severe myocardial failure such as one sees after pulmonary infarction caused sudden increase in the bilirubin content of the blood, we cannot accept the view that jaundice that develops under these circumstances is caused primarily by the absorption of locally formed bilirubin. We do not mean to imply, however, that increased blood destruction plays no rôle in the development of "cardiac" jaundice. The evidence that there is increased blood destruction and bilirubin formation in myocardial insufficiency was fairly well established (Eppinger, 1923). One must conclude, therefore, that since the ability of the liver to excrete bilirubin is already impaired in myocardial failure, the degree of bilirubin retention in the blood stream must be enhanced by the abnormally large amount that the liver must deal with. It is possible, perhaps, that the bilirubin that is formed locally about an infarct may increase to some extent the amount of bilirubin that accumulates in the blood. It has been our purpose, however, to show that the absorption of bilirubin formed locally about an infarct is not the *primary* cause.

4 *Impairment of bilirubin formation* We do not know the agency which forms bilirubin. It is obvious that under different conditions bodily functions are carried out at different levels of efficiency, and in jaundice one must naturally consider the matter of the bilirubin formed under those conditions. We have seen, in the experiment on dog 4

(table 4) that the bilirubin content of the blood may decrease even though there is every reason to believe that the liver damage is increased, and in the experiment on dog 6 it was found that the explanation probably lay in the diminution in the amount of bilirubin that was formed. Whether this diminution in bilirubin formation was actually due to impairment of the function of the agency which forms bilirubin, we do not know. Such a possibility exists, however, and although it is rarely of great significance, it must be taken into account if every factor concerned with the cause of jaundice in myocardial insufficiency is reckoned with. We have observed in patients as well as in experimental animals a discrepancy in the amount of bilirubin actually in the blood and that which we expected to find. The following case is an instance.

A woman with severe myocardial insufficiency and pronounced jaundice improved temporarily under the usual therapeutic measures. Following an attack of acute tonsillitis, she became rapidly worse and after several weeks died of progressive myocardial failure. Toward the end there was generalized infection with streptococcus hemolyticus. In spite of the fact that myocardial failure was extreme, and the liver at necropsy showed extensive necrosis, the bilirubin content of the blood was only 0.8 unit a few days before the death of the patient.

5. Impairment of renal function. We have not actually investigated the relation between renal function and the development of bilirubinemia. It is well known that renal function may be disturbed in myocardial failure, and that occasionally the impairment may reach a considerable degree (Resnik and Keefer, 1925). It is conceivable that this damage may play a rôle in favoring the accumulation of bilirubin in the blood because of the inability of the kidney to excrete it properly. But, since patients with severe myocardial insufficiency and jaundice excrete large amounts of bilirubin, it does not seem probable that a single factor such as this should play a very important rôle in determining the amount of bilirubin in the blood.

6. Mechanical obstruction of the bile passages. Eppinger (23) has described the occurrence of so-called bile thrombi in the liver of chronic passive congestion, and has suggested that these cause a certain degree of obstructive jaundice. Eppinger admitted, however, (1923) that obstructive jaundice caused by bile thrombi could not be the sole,

nor even an important factor in bringing about jaundice in myocardial failure, since bile thrombi are found usually only with difficulty or are entirely absent, even though jaundice is present.

7 *Hematinemia* Schottmüller (1914), Thormählen (1918) and others have described cases, of extravasation of blood into the tissues, including cases of pulmonary infarction, in which jaundice was due apparently to the presence of hematin in the blood serum. In the cases of jaundice following pulmonary infarction that we have observed, there has been practically always associated bilirubinuria, in two cases, it was shown that the bilirubin content of the blood increased markedly at the time jaundice appeared. In our cases, then, one can account for the development of jaundice purely by the increased bilirubin content of the blood. There may be other cases, however, in which jaundice is due to hematinemia, the differentiation of these cases from those in which bilirubin is the jaundice-producing pigment must be made by the van den Bergh test and spectroscopic examination of the blood.

We have discussed in considerable detail the various factors that may be at work in the production of jaundice in myocardial insufficiency, and in some measure their probable importance. We wish particularly to stress the conception that anoxemia may be of great significance in these, and undoubtedly in other cases of jaundice. We do not propose to discuss in detail, at this time, the relation of anoxemia to these other instances of jaundice, it will suffice to mention one, namely, that occurring in lobar pneumonia. This disease is occasionally associated with conspicuous jaundice, and more frequently with latent jaundice (Lepehne, 1921) (Eppinger, 1923). Histological examination of the liver in fatal cases usually shows the changes that are commonly found in other infectious diseases, namely, cloudy swelling of the parenchymal cells. While it is true that there is increased blood destruction in this disease, particularly in the lungs, it is highly probable that anoxemia, which is usually present and which often may be extreme, plays an important rôle in reducing the function of the liver.

SUMMARY

In a group of animal experiments, it has been demonstrated that anoxemia may depress the function of damaged livers to such an extent

that bilirubinemia appears. Reasons were given for assuming that pulmonary infarction in patients with myocardial insufficiency produces anoxemia, probably severe in some cases, and it was chiefly to increased anoxemia that we ascribed the sudden appearance of jaundice in these patients. It was also shown that anoxemia failed to produce bilirubinemia when the liver was normal and when anoxemia, even though severe, was present for several hours only. When uninjured livers were however, subjected to anoxemia, histological examination showed beginning central necrosis of the lobules. The suggestion was made, therefore, that not only does anoxemia depress the function of the liver, when already damaged, but that anoxemia, if it is of sufficient severity and long standing, may actually cause, in part at least, the anatomical changes which one sees in chronic passive congestion. The various factors, and their probable importance were discussed, that may play a rôle in "cardiac" jaundice, particularly with reference to that which appears after pulmonary infarction. Finally, it was pointed out that anoxemia may be of importance in causing jaundice that is associated with other diseases, such as lobar pneumonia.

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THE NUMBER OF FORMED ELEMENTS IN THE URINARY SEDIMENT OF NORMAL INDIVIDUALS*

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(Received for publication February 23, 1926)

The general methods which for so long have been used in counting the cells of the blood are applicable in the enumeration of the formed elements of the urine. But in urine as opposed to blood there are such wide variations in volume, salt concentration and reaction that it is only under certain special conditions that quantitative results can be obtained. Thus, in order to eliminate the effect of changes in volume the urine must be collected over a measured interval of time, so that the number of formed elements excreted per unit of time may be determined. Rates of excretion, not concentrations, are essential, for otherwise it would be impossible to tell whether a change from, say, 200 to 100 in the number of cells per cubic centimeter of urine was due to a decrease in the number of cells or to an increase in the volume of urine. Again, certain changes in salt concentration and reaction may lead to the partial or complete disappearance of some of the formed elements of the urine. In dilute and alkaline urine red blood cells may be so completely lysed that they cannot be counted, and hyaline casts dissolve. A pronounced decrease in the number of casts in the urine of patients with Bright's disease may be readily brought about by simply increasing the volume of urine by giving water, since the hyaline casts and all casts with a hyaline matrix disappear when the urine becomes dilute. It has also been noted that a certain acidity, the degree of which depends on the salt concentration of the urine, is necessary for the preservation of hyaline casts. In test tube experiments it has been found that the *minimal* concentration of NaCl required to keep suspensions of washed hyaline casts from

* This work was aided by the Wellington Gregg Fund for the investigation of Bright's disease.

dissolving increases as the H-ion concentration is decreased. These more or less qualitative observations are sufficient to indicate that reliable counts can be secured only in collections of urine in which a certain degree of concentration and acidity is attained.

The particular means adopted to meet the requirements outlined above, the mode of urine collection and preservation, and the technique used in counting, have already been fully described elsewhere (1). In this paper are given the averages and ranges of variation in the twelve-hour rates of excretion of casts, red blood cells and of white cells and epithelial cells found in urine obtained from normal individuals.

The subjects were all working medical students who led a quite uniform and physically inactive existence. The collections were started at about 8:30 in the evening and the whole night urine was collected at one voiding twelve hours later when they came to work.

TABLE 1

The rate of excretion per twelve hours of formed elements in the urine of normal men
Averages from 82 counts on 74 individuals

Casts	Red blood cells	White blood and epithelial cells
1,040	65,750	322,500

in the morning. No fluids of any sort were taken after breakfast on the preceding day and as a consequence the twelve-hour volume was as a rule quite small. The average amount was 352 cc. The average specific gravity was 1.031.

Table 1 gives the average number of casts, red blood cells, and white blood and epithelial cells found in 82 examinations of 74 individuals. In table 2 the counts are arranged in order of magnitude.

The average number of casts found in each twelve-hour collection was 1040. An inspection of table 2 shows that casts were found in 45 of the 74 individuals and were noted as absent in 29. But a consideration of the method employed makes it quite apparent that neither the figures given in those instances in which casts were discovered nor the zeros entered in the remainder can be regarded as having any individual quantitative significance. In the urine of patients with Bright's disease in which hundreds of thousands or millions of casts

TABLE 2

Ranges of variation in the rates of excretion per twelve-hour night period of casts, red blood cells and white and epithelial cells in the urine of apparently healthy men

Casts	Red blood cells	White and epithelial cells
4,270	425,000	1,835,000
4,230	422,000	1,230,000
4,140	417,000	859,000
3,150	353,000	773,000
2,810	276,000	724,000
2,695	270,000	686,000
2,340	237,000	668,000
2,240	208,000	651,000
2,220	194,000	628,000
2,220	187,000	616,000
2,220	181,500	560,000
2,220	162,000	480,000
2,170	159,000	471,000
2,060	145,000	456,000
1,970	118,000	438,000
1,780	104,000	408,000
1,780	74,000	395,000
1,690	73,000	372,000
1,690	70,000	364,000
1,640	69,800	360,000
1,535	63,400	359,000
1,450	56,400	350,000
1,400	53,000	349,000
1,390	45,500	345,000
1,380	38,500	322,200
1,370	32,200	315,000
1,350	29,800	300,000
1,350	26,500	272,000
1,290	26,100	271,000
1,260	22,200	265,000
1,210	19,400	259,000
1,200	16,100	259,000
1,180	10,100	244,000
1,159	9,860	239,000
1,110	5,060	233,000
1,090	3,100	228,000
1,070	0	210,000
1,050	0	206,000
1,050	0	203,000
925	0	202,000
840	0	196,000
704	0	182,000

TABLE 2—*Continued*

Casts	Red blood cells	White and epithelial cells
676	0	174,000
658	0	172,000
645	0	168,000
583	0	165,000
580	0	163,000
556	0	158,000
528	0	157,000
0	0	147,000
0	0	142,000
0	0	139,000
0	0	130,000
0	0	130,000
0	0	129,000
0	0	126,000
0	0	120,000
0	0	119,000
0	0	116,300
0	0	113,400
0	0	111,000
0	0	90,800
0	0	84,000
0	0	80,400
0	0	66,800
0		60,600
0		56,400
0		40,000
0		32,400
0		
0		
0		
0		

are excreted every twelve hours the numerical results are trustworthy, but in these normal urines never more than 5 and often only 1 cast was seen in any one collection. It therefore seems quite likely that a more laborious search would have disclosed some casts in most if not in all of those specimens which are reported here as containing none¹. But though no reliance can be placed on any single figure of

¹ The total ruled area of 9 sq mm in the ordinary type of blood counting chamber was counted over from 4 to 10 times with separate drops of sediment. In the majority of cases 8 drops were used. This represents a volume of 0.0072

the series, the average result is probably a close approximation to the number of casts excreted by normal individuals when the conditions permit the formation and preservation of these structures

The range of variation was from none to 4270 casts in twelve hours, and though here again no stress can be laid on the actually observed limits, they at least define the order of magnitude in the number of casts to be expected in the urine from normal individuals, and I have found this estimate of value in determining the significance of the sediment results in patients in whom some very minor renal lesion was suspected. It was of course for this reason that counts were made. The fact that urine from healthy individuals may contain casts was established by Haines and Skinner in 1898 (2)

The type of cast found was the hyaline variety. There were only three instances in which a single epithelial or granular cast was discovered. The hyaline casts only rarely contained any cell inclusions and fat droplets were never seen.

The average number of red blood cells was 65,700 per twelve-hour period. It seems to be generally assumed that the presence of red cells in urine is always indicative of some abnormality in the urinary tract, but this view is certainly erroneous. On the other hand they are probably not constant urinary constituents for I have sometimes failed to find them in spite of prolonged search in normal individuals in whose urine they had previously been present. They were discovered in 40 of the 64 examinations made, and might very well have been found in a larger percentage if more time had been spent in searching for them. However the main interest lay in the determination of the range of variation. This was found to extend from none to 425,000 per twelve-hour period, a fact of special importance in the diagnosis of the latent stage of haemorrhagic nephritis. It is clear from the above figures that it is only when the red blood cells are excreted at a rate of a million or more in each twelve hour period that they can be regarded as significant. However it should be remembered that without some experience and a good deal of care

cc. The sediment was concentrated by centrifuging from a 10 cc. sample of the mixed urine to a volume which varied from 0.1 to 0.3 cc. but usually 0.2 cc. With a count through 0.0072 cc. from a sediment of 0.2 cc. and a urine volume of 352 cc., the fraction of the total urine actually examined represents only 1 part in 972.

one is apt to pass over many of the forms which red blood cells may assume in urine. Particularly in the urine of patients they may be fragmented or almost wholly lysed, so that only faint shadows remain which are easy to miss.

The average number of white blood cells and of small round epithelial cells in the twelve hour out-put of urine was 322,500 and the range of variation lay between 1,835,000 and 32,400. Both white blood and epithelial cells were counted together because it was frequently impossible to decide to which of these two classes a given cell belonged. Of course, from the standpoint of renal diagnosis, what is really wanted is a method by which those cells which come from the kidney can be distinguished from all others. A great many stains were tried but without success and in the end it was concluded that size was the only criterion. The large cells from the surface layers of the mucous membrane of the urinary tract were not included and the medium sized cells which originate from deeper layers were passed over when it was considered that their diameter was greater than that which could well be attained by even a swollen renal cell. This is admittedly a vague method of differentiation and one under which a number of cells from the prostate and other glands of the urinary tract are included. The wide range of variation in the counts is indeed in all probability to be accounted for largely by the inclusion of latent cases of prostatitis and urethritis. In practice however it has not proved an unsatisfactory standard because the number of white blood and epithelial cells found in the urine of patients in whom an active renal lesion exists is great enough to far surpass our upper limits of normality.

CONCLUSIONS

The rate of excretion of casts, red blood cells, and white blood and epithelial cells was determined in 74 medical students under conditions favorable to the preservation of these urinary constituents. The following results were obtained

Rate of excretion per twelve-hour period

	Average	Lowest	Highest
Casts	1,040	0	4,270
Red blood cells	65,750	0	425,000
White blood and epithelial cells	322,500	32,400	1,835,000

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- 2 Hanes and Skinner Jour Amer Med Assoc., 1898, xxx, 234

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CONCLUSIONS

The rate of excretion of casts, red blood cells, and white blood and epithelial cells was determined in 74 medical students under conditions favorable to the preservation of these urinary constituents. The following results were obtained

THE EFFECT OF SOME PHYSIOLOGICAL VARIABLES ON THE NUMBER OF CASTS, RED BLOOD CELLS AND WHITE BLOOD CELLS AND EPITHELIAL CELLS IN THE URINE OF NORMAL INDIVIDUALS*

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(Received for publication March 8, 1926)

In the course of clinical work with patients suffering from Bright's disease certain questions constantly present themselves which as constantly are left unanswered except by practical decisions of an almost completely arbitrary nature. For instance, how long should a patient be kept in bed when the initial stage of haemorrhagic Bright's disease has passed, when the edema has gone and the blood pressure has returned to its usual level? Certainly until examinations of the urinary sediment indicate a subsidence in the first intensity of the inflammatory process in the kidney, but also quite certainly not until all signs of inflammation have disappeared, since the latent stage of the disease extends over months or years or decades and during all this time the urine indicates the continuance of an active renal inflammation. If a pronounced increase in the number of casts, red blood cells and white blood cells and epithelial cells were observed after the patient was first out of bed, we might feel fairly certain that further rest was necessary, but it is impossible to interpret the significance of a moderate or slight increase. We know nothing about the response of the normal kidney to like conditions. It may be that some degree of increase in casts and red blood cells is the natural reaction of the kidney to the slight jars and jolts which it receives with every movement of the body. Again in the treatment of the actively inflammatory types of Bright's disease it has for long been the custom to reduce the quantity of protein in,

*This work was aided by the Wellington Gregg Fund for the investigation of Bright's Disease

differences observed might well have been the result of chance and we must conclude that no appreciable difference has been shown to exist between the day and night sediments

II EFFECT OF VIBRATION

Since the ordinary day time activities were without effect, another experiment was carried through in which a more directly traumatic influence on the kidney was introduced. The subjects sat on a chair

TABLE 2
The effect of shaking on a vibrating chair
Averages from a group of 9 individuals

Casts per 12 hours		Red blood cells per 12 hours		White and epithelial cells per 12 hours	
No shaking	Shaking	No shaking	Shaking	No shaking	Shaking
582	378	70,700	212,100	313,000	587,000

TABLE 3
The rate of red blood cell excretion without and with shaking

Name	No shaking per 12 hours	Shaking per 12 hours
B	87,000	291,000
H.	120,000	144,900
W	27,000	202,800
M	44,400	291,000
McB	0	56,400
A.	0	0
S	237,900	558,000
G	66,000	30,000
N	54,000	336,000

which rested on steel springs and which was shaken by a motor. In this manner strong vibrating movements were transmitted to them not unlike those experienced in riding on a motor-bicycle. Half an hour was as much as could be tolerated without too much discomfort. Collections of urine were made from 8.30 a.m. to 12.30 p.m. after abstention from fluids during the preceding day. The shaking extended from 8.30 a.m. to 9.00 a.m. The average results expressed as 12-hour rates are given in table 2.

the food to a minimum, because the work of the kidney is thereby lessened and rest has been found efficacious in the treatment of inflammation in general. However there are special reasons for hesitancy in accepting conclusions drawn by analogy from observations on other organs as necessarily true for the kidney, and in actual fact the effects of protein on the kidney are still the subject of experiment and disagreement. Direct evidence might be obtained by observing the effect of large amounts of protein on the urinary sediment of patients with Bright's disease but a prerequisite is a knowledge of the changes which may thus be produced in the urine of normal individuals. An attempt was therefore made to estimate the influence on the urinary sediment of medical students of certain types of bodily movement and also to obtain a measure of the effect of the administration of a large quantity of protein.

TABLE 1

The rate of excretion of formed elements during the day compared with the rate of excretion during the night

Averages from a group of 18 individuals

Casts		Red blood cells per 12 hours		White and epithelial cells per 12 hours	
Day	Night	Day	Night	Day	Night
852	827	48,900	65,600	434,000	259,000

I DAY AND NIGHT URINES

A group of 18 students collected urine from 8 30 p m to 8 30 a m (night urine) and from 8 30 a m to 12 30 p m (day urine). In both cases the results are expressed as 12-hour rates. No fluids were taken for 24 hours so that the urine might be concentrated. The night urine had an average volume of 382 cc with a specific gravity of 1 029. It is of interest that the day volume, though secreted during the last 4 hours of the period of abstention from fluids, had a larger volume—522 cc per 12-hour period with a specific gravity of 1 029. The average counts are given in table 1.

On account of the great variability in the rates of excretion of the various formed elements of the urine the difference between group averages must be large to be significant. In this instance the actual

A simple inspection of table 5 is enough to indicate that there is no proof that the meat was the cause of the observed increase in the average rate of red blood cell excretion, for that increase is accounted for in the main by an unusually large excretion in a single individual. This impression is confirmed by statistical analysis. The "probable difference between the averages" is 205,000 while the actually observed difference is only 120,000.

TABLE 5
The rate of red blood cell excretion without and with meat

Name	No meat per 12 hours	Meat per 12 hours
H.	7,300	0
G	0	0
W	1,500	0
B 1	0	0
B 2	0	61,000
B 3	0	34,000
D	2,600	0
M	182,000	442,000
Be.	0	0
Da.	0	510,000
Gr	4,200	1,190,000
Bu.	276,000	27,200
Bur	0	0
K.	0	119,000
S	208,000	63,200
Ga	56,400	272,000
N	22,300	89,000

CONCLUSION

Quantitative determinations of the number of formed elements in the urine of normal individuals failed to show that either bodily movements of various types or the ingestion of a large amount of protein in the form of meat had any statistically significant effect.

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The greatest difference is a threefold increase in the number of red blood cells after shaking. The individual counts (table 3) show a fairly well distributed increase after shaking and at first glance it would seem that a quite definite effect had been produced. But the "probable difference between the averages" of the two series is not much less than the actual difference, and this result might therefore be a chance. It may accordingly be concluded that vibration did not lead to a significant increase in any of the formed elements in the urine.

III THE EFFECT OF THE ADMINISTRATION OF A LARGE AMOUNT OF MEAT

A group of 25 students took $1\frac{1}{2}$ pounds of steak at 6 30 p m and collected urine from 8 30 p m until 8 30 a m. No fluids were taken

TABLE 4
The effect of the administration of a large amount of meat
Averages from a group of 25 individuals

Casts per 12 hours		Red blood cells per 12 hours		White and epithelial cells per 12 hours	
No meat	Meat	No meat	Meat	No meat	Meat
1,078	681	45,000	165,000	309,000	244,000

after 12 30 p m of the day preceding the completion of the collection. The average results are compared with the control tests in table 4. In the control experiments the average volume was 378 cc with a specific gravity of 1 030. After meat the volume was 479 cc with a specific gravity of 1 033.

The only difference which might possibly have a meaning is the increase in the number of red blood cells after eating the meat. Unfortunately, however, the red blood cells could be counted in only 17 of the 25 observations after meat, because in the remainder there was a deposit of phosphates in the urine. Instead of making the urine more acid, as might have been expected, the ingestion of the meat was followed in these instances by the secretion of an alkaline urine. The fact that meat often causes the urine to become alkaline has already been noted by Lutz (1). The individual data in these 17 comparisons are given in table 5.

MEASUREMENTS ON THE SKIN CAPILLARIES IN CASES OF POLYCYTHEMIA VERA AND THE RÔLE OF THESE CAPILLARIES IN THE PRODUCTION OF ERYTHROSIS

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INTRODUCTION

One of the outstanding features of polycythemia vera is the color of the skin. Clinical descriptions employ the terms brick red, congestive facies, cyanosis and erythrosis. Vaquez, who reported the first observed case of this disease, noted cyanosis as the prevailing tint. Osler thought that the color change was not that of cyanosis but congestion or redness. Since the microscopic investigation of the skin capillaries has shown definite and measurable variation of these vessels in this disease, it seemed to us that a quantitative analysis of the blood and vascular factors would add much to an adequate understanding of the color changes, and herewith report the results of such analysis.

The color of the skin in cases of polycythemia vera is predominately red but subject to marked variations. The redness affects largely the facial and acral regions, the hands to the glove line, and the face and neck to the décolleté margin. The feet are usually less red than the hands. The mucous membranes appear cyanotic when the skin is definitely red. Cyanosis has not been observed in the conjunctiva, this tissue has a bright red, or an injected or inflamed appearance. On the trunk and proximal portions of the extremities color changes are much less than on the more distal and facial areas. The skin over the trunk and back may have the mild flushed appearance of a scarlatiniform rash.

Clinical descriptions of the color of the skin in our cases by various co-workers were "markedly red," "brick red," "reddish cyanosis," and

The amount of the oxygen unsaturation of the capillary blood determines the presence of cyanosis. Lundsgaard and Van Slyke have shown that a normal range of venous oxygen unsaturation in a patient with polycythemia vera is 5 to 6.6 per cent by volume. Cyanosis becomes visible in the skin when the mean oxygen unsaturation of the capillary blood is in excess of 6 per cent by volume. They attribute the marked tendency toward cyanosis to two conditions: the increased content of hemoglobin, which magnifies the existing oxygen unsaturation, and the abnormal condition of the capillaries as relating to changes in number and width. The rate of capillary flow is probably an important factor.

The part played by the superficial venules in producing the color of the skin is not known. Goldschmidt and Light have shown a type of cyanosis, not related to oxygen unsaturation and probably due to increased peripheral venous pressure.¹ This form of cyanosis is observed with the hand in the pendent position and is relieved by elevation.

METHODS OF INVESTIGATION

Studies were carried out to determine (1) the average number of capillary loops for each square millimeter of skin in normal persons and in patients with polycythemia vera, (2) the relationship of the number and area of open capillaries to the total blood and hemoglobin volumes, and (3) the ratio of the total area of exposed capillary blood to each square millimeter of skin surface.

Five patients presenting the typical clinical syndrome of polycythemia vera were observed. The spleen and liver were enlarged in all. The blood volume, number of erythrocytes and percentage of hemoglobin showed the large increases typical of this disease.

The total circulating cell, plasma and hemoglobin volumes were determined by the dye method, according to the technic described by Keith, Rowntree, and Geraghty, except that Congo-red instead of vital red dye was employed. The total circulating hemoglobin was determined by multiplying the total circulating blood volume by the grams percentage of hemoglobin. The hemoglobin was determined by

¹ We have obtained increased values for oxygen unsaturation of the arterial blood in one case.

“markedly cyanotic” Observation of the color by different or by the same observers have shown variations during the same, and on different, days Redness is common during the warmer months, and cyanosis during the colder months In the cool period of early morning, cyanosis is more marked, and at mid-day, less marked Age and sex seems to play a part Younger persons are less cyanosed than older ones, and it seemed as if females were less cyanotic than males In brief, the color of the skin in cases of polycythemia vera in the acral and facial areas, is that of erythrosis, with a marked tendency toward cyanosis

COMPONENTS OF SKIN COLOR

The color of the normal Caucasian skin is a complex composite of several factors, and subject to many variations incident to environmental, psychic, and postural influences In attempting to differentiate the various components the following are the most important data

The major factor is the blood content of the vessels of the skin, which is affected by the following variable factors (1) the absolute amount of circulating cells and hemoglobin, and (2) area of exposure of the capillary blood, which depends on the number of open capillaries for each unit of surface area and the area of the exposed portion of the capillary loops, the minor factors are (1) the thickness and pigment content of the epidermis, (2) the color of the plasma, as in jaundice, (3) variations in the oxygen unsaturation of the capillary blood, and (4) color effects of the superficial venules

For estimating the relative importance of the major and minor factors, very little information is available other than that from the blood There is no available information on the capillaries, and the thickness and color values of the epidermis, the latter of which may become of major importance, as in Addison's disease Pigmentation is not an outstanding features of polycythemia vera, although the effect of weather exposure with tanning undoubtedly is of some importance in determining the areas of greatest erythrosis

Quantitative estimations of the color of the plasma can be obtained by determining the bilirubin content of the blood serum When this substance has a concentration of about 5 mg or more for each 100 cc of plasma, jaundice is recognizable in the tissues

TABLE 1

*Measurements of the capillaries of the skin in cases of polycythemia vera and of controls**

Case	Age	Sex	Clinical diagnosis	Region examined	Average area of blood visible in capillary † sq. mm	Number of capillaries visible in each square millimeter	Ratio of area of capillary blood exposure unit (1 sq. mm.) skin area, per cent
1	57	M	Polycythemia vera	{ Nail fold First joint Second joint	0.0038 0.0007 0.0020	45 80 56	17.0 5.6 11.2
2	62	M	Polycythemia vera mild essential hypertension	{ Nail fold First joint Second joint	0.0033 0.0008 0.0044	52 85 46	17.2 6.8 20.2
3	58	M	Polycythemia vera, severe hypertension	{ Nail fold First joint Second joint	0.0041 0.0021 0.0030	40 70 65	16.4 14.7 19.5
4	37	F	Polycythemia vera	{ Nail fold First joint Second joint	0.0036 0.0013 0.0018	42 65 88	15.1 8.5 15.8
5	52	M	Polycythemia vera	{ Nail fold First joint Second joint	0.0052 0.0012 0.0048	38 108 50	19.8 12.9 24.0
Average values					0.0028	62	15.0
	26	M	Normal	{ Nail fold First joint Second joint	0.0051 0.0007 0.0008	25 38 50	7.7 2.7 4.0
	32	F	Normal	{ Nail fold First joint Second joint	0.0021 0.0005 0.0009	42 40 53	8.4 2.0 4.8
Average values					0.0013	41	4.9

* All data are from the right hand

† Five to 10 areas averaged in each

The ratio of the area of the capillary blood exposure to a unit area of skin (1 sq. mm.) was calculated

A slight error exists in this type of investigation, because of the

the acid hematin method, using the permanent standard of Haden² The nail-fold capillaries were examined by the Lombard method The technical details have been described in former papers Instantaneous photomicrographs (Sheard's technic) of the capillaries, the nail-fold vessels and skin over first and second joints were made (figs 1 and 2) The films were projected on a screen and a total magnification of 750 diameters was obtained The irregular areas,



FIG 1



FIG 2

FIG 1 PHOTOMICROGRAPH OF SKIN CAPILLARIES AT THE NAIL-FOLD IN A YOUNG NORMAL SUBJECT³

Note the collecting venules and capillary bed ($\times 25$) Time of exposure one-tenth second

FIG 2 PHOTOMICROGRAPH OF CAPILLARY TOPS IN THE SKIN IN THE FIRST JOINT IN THE SAME PERSON AS IN FIGURE 1 ($\times 25$)

Time of exposure one-eighth second

representing the capillary areas or exposed capillary blood, were traced on paper and the dimensions obtained by the use of a planimeter

The total area of the exposed capillary blood was computed by obtaining the average of ten such areas and multiplying by the average number of open capillaries for each square millimeter of skin area

² Fifteen and six-tenths grams of hemoglobin for each 100 cc of blood was taken as the normal, 100 per cent

³ All photomicrographs reproduced in this paper are made from the original untouched negatives.

any marked variation in the number of capillaries in the same subject. The conditions of the examination were quite comparable.

An average of 41 loops (range 25 to 53) for each unit area of skin was found in the normal subjects (table 1). The average area of the capillary summit, or the area of exposed capillary blood, in the patients

TABLE 3

The effect of the reduction of the blood on the area and number of capillaries in cases of polycythemia vera

Case	Date	Hemoglobin		Circulating blood volume cc	Circulating hemoglobin g/m ¹⁰⁰	Region examined	Average area of capillary blood visible in loop in square millimeter	Capillaries visible in each square millimeter of skin	Ratio of area of capillary blood exposure to unit skin area per cent
		Grams per cent	Percentage						
4	6 29 25	19.0	125	7,700	1,490	Nail fold	0.0036	42	15.1
						First joint	0.0013	65	8.5
						Second joint	0.0018	88	15.8
						Average	0.0023	65	13.1
	7 11 25	8.7	57	4,380	380	Nail-fold	0.0028	30	8.4
						First joint	0.0012	26	3.1
						Second joint	0.0018	46	8.3
						Average	0.0019	30	6.6
5	4 28 25	23.4	153	11,000	2,590	Nail fold	0.0041	40	16.4
						First joint	0.0021	70	14.7
						Second joint	0.0030	65	19.5
						Average	0.0031	65	16.8
	5 20, 25	9.5	62	5,050	630	Nail fold	0.0024	32	7.7
						First joint	0.0009	45	4.1
						Second joint	0.0012	36	4.3
						Average	0.0015	37	5.3

with polycythemia vera was 0.0028 sq. mm. This is more than twice the area (0.0013) found in the normal subjects.

The ratio of the total area of exposure of the capillary blood to each square millimeter of skin surface averaged 15 per cent in the cases of polycythemia vera (table 1). This is three times greater than for

shifting of the number of open capillaries in the skin and irregularities in the areas measured. A series of comparative photomicrographs of the same area taken over a short period of time under controlled conditions showed only a slight variation in the number of open vessels. There are also a number of capillary loops in the deeper layer of the epidermis which are not visible but probably play a part in determining the color of the skin. The venules likewise contribute to the color, but we could not convince ourselves just how important they are.

TABLE 2
Blood and capillaries in cases of polycythemia vera

Case	Hemoglobin		Erythrocyte millions	Blood		Plasma		Total circulatory hemoglobin in grams	Average number of capillaries visible each square millimeter skin	Ratio of area of capillary blood each square millimeter skin*	Color of skin
	Grams per cent	Percentage		Volume cc.	Cc for each kilo gram	Volume cc	Cc for each kilo gram				
1	26.0	171	7.00	14,700	180	3,670	46	3,700	62	12	Marked erythrosis with cyanosis
2	24.5	161	5.02	8,580	136	3,430	54	2,100	61	15	Mild erythrosis
3	31.1	195	7.00	12,400	213	3,600	62	3,850	98	19	Marked erythrosis and cyanosis
4	19.0	125	6.90	7,700	167	2,250	48	1,490	65	14	Mild erythrosis
5	23.4	153	6.12	11,000	173	5,300	52	2,590	60	17	Moderate erythrosis
	15.6	100	4.50	5,100†	85	3,000†	50	800	41	4.9	Average normal values

* Per cent

† Calculated for a person with body weight of 60 kg

On the microscopic examination of the skin of many patients the color between capillaries is very light, whereas that of patients with other diseases, especially polycythemia vera, shows a pinkish tint in the deeper layers. There is no doubt, however, that the greatest amount of color is contributed by the capillary blood.

The intensity of the cyanosis varied according to the environmental temperature. The average number of open capillaries in cases of polycythemia vera varied from 38 to 108 for each unit area of skin, averaging 62 (table 1). We were not able to demonstrate

normal subjects (4.9 per cent). The area of greatest capillary blood exposure was found in the nail-folds and the skin near the second joint.

There is no constant relationship or parallelism between the degree of increased volume of the blood and the number of open capillaries (table 2). The relationship was fairly constant between the area of exposed capillary blood and the blood and hemoglobin volume. In Case 3 is shown the greatest increase in the volume of the blood per

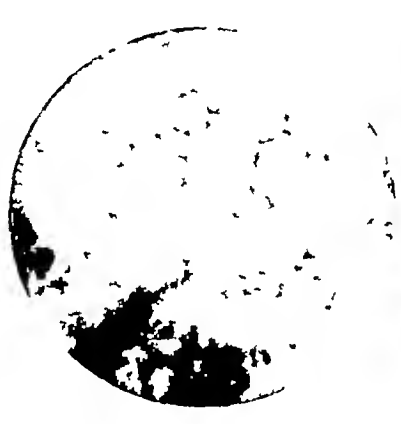


FIG 7

FIG 7 PHOTOMICROGRAPH OF THE CAPILLARY TOPS IN CASE OF POLYCYTHEMIA VERA, SKIN AREA IN REGION OF FIRST JOINT, BEFORE TREATMENT $\times 25$

Time of exposure one-tenth second



FIG 8

FIG 8 PHOTOMICROGRAPH OF THE CAPILLARY TOPS IN CASE OF POLYCYTHEMIA VERA, SAME SKIN AREA AND SAME FINGER AS IN FIGURE 7, AFTER TREATMENT $\times 25$

Time of exposure one eighth second

kilogram and hemoglobin with the greatest number of open capillaries and the greatest ratio of capillary blood exposure to each square millimeter of skin.

The capillary and blood factors before and after reduction of the blood volume by treatment with phenylhydrazin have been studied (figs 3, 4, 5, 6, 7 and 8). In Case 4 (table 3), the ratio of the total area of exposed capillary blood to each square millimeter of skin was 13.1 per cent and the average area of visible capillary blood in each

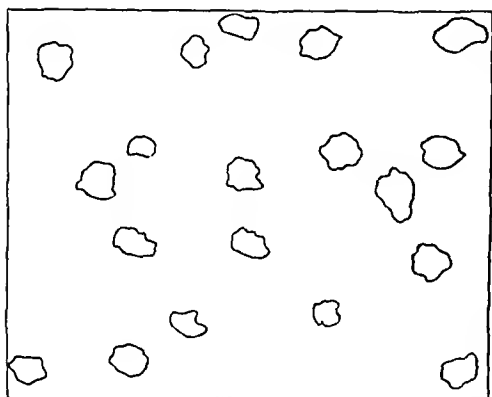


FIG 3

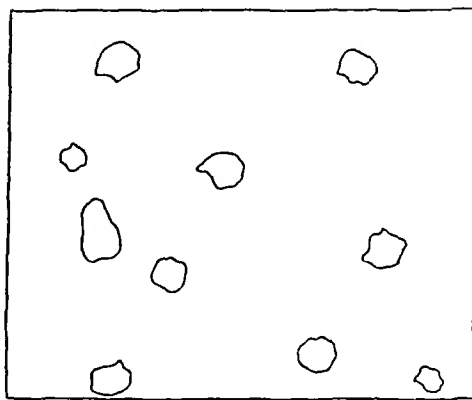


FIG 4

FIG 3 SKETCH OF BLOOD AREAS IN CAPILLARY TOPS IN A CASE OF POLYCYTHEMIA VERA, FIRST JOINT, BEFORE TREATMENT $\times 750$ (Reduced $\frac{1}{3}$)

FIG 4 SKETCH OF BLOOD AREAS IN CAPILLARY TOPS IN THE SAME REGION OF THE SKIN AS IN FIGURE 3, AFTER TREATMENT $\times 750$ (Reduced $\frac{1}{3}$)



FIG 5



FIG 6

FIG 5 PHOTOMICROGRAPH OF THE SKIN CAPILLARIES AT THE NAIL-FOLD IN CASE OF POLYCYTHEMIA VERA, BEFORE TREATMENT $\times 25$

Time of exposure one-tenth second

FIG 6 PHOTOMICROGRAPH OF THE SKIN CAPILLARIES AT THE NAILFOLD IN CASE OF POLYCYTHEMIA VERA, SAME SKIN REGION AND SAME FINGER AS IN

FIGURE 5, AFTER TREATMENT $\times 25$

Time of exposure one-tenth second

The increase in the capillary measurements exceeds the increase in the blood elements, thus the capillary factors in the production of erythrosis are apparently the most important, although their variations are of course dependent on the augmentation of the blood volume

The response of the capillaries to the reduction in the cell volume following treatment varied somewhat in the two cases studied. In both, the greatest change occurred in the number of open capillaries, which was reduced about 50 per cent.

It is difficult to determine the importance of the venules in the production of erythrosis in cases of polycythemia vera. These vessels are not clearly visible but contribute somewhat to the color of the skin as the tissue has a pinkish tinge between the capillary loops. The superficial venules of the cheek and nose are dilated in a few patients, especially those subject to exposure to the wind and sun. The areas of venous dilatation show cyanosis to a marked degree, as would be expected.

SUMMARY

The large increase in the circulating volume of erythrocytes in cases of polycythemia vera produces distention of the blood vessels, the venous vessels assuming the larger portion of this increase. The capillaries of the skin become distended, more markedly in their venous portion, and additional capillaries are formed. These changes have been studied quantitatively by photomicrographic methods, and compared to similar data obtained from normal subjects. The ratio of the total area of visible capillary blood to a unit area of skin is determined and found to be definitely increased. The demonstrable increase in the size and number of the capillaries exceeds the changes in the blood. There is a gradual decrease in the size and number of capillaries with reduction of the cell volume.

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loop was 0.0023 sq. mm. The average number of loops was sixty-five for each square millimeter of skin area. The blood volume was reduced to 4,380 cc. and circulating hemoglobin to 380 grams. The ratio of the area of the exposed capillary blood to a unit area of skin diminished to 6.6 per cent and the average number of loops to thirty, while the average area of the visible capillary blood diminished to 0.0019 sq. mm. In Case 5 (table 3) the blood volume was reduced by treatment. The ratio of the total area of the exposed capillary blood to unit area of skin decreased from 16.8 to 5.3 per cent. The number of loops and the average area of the visible capillary in each loop decreased one-half.

DISCUSSION

The color of the skin in cases of polycythemia vera is due largely to one primary factor, the large increase in the circulating cells and hemoglobin, and two modifying factors, the number of capillaries for each unit area of skin and the area of capillary blood exposed in each capillary. The circulating cell volume, according to the dye method, is increased approximately 75 to 100 per cent above the average normal. As a result of the large increase in the blood volume the capillary bed of the skin assumes a storage function. As has been shown, this involves chiefly the venous segment, which portion extends well over on the arterial side of the loop. Proximal to the nail-fold area the skin capillaries are arranged at right angles to the skin surface, so that only the summits or crests of the loops are visible but this portion is distended and its area is measurably enlarged. The area of the capillary crest was approximately double that in normal subjects.

Further accommodation for the increase in blood volume is made by an increase in the number of open capillaries. Our data show an average of sixty-two loops for each square millimeter of skin area in cases of polycythemia vera, as compared to forty-one in normal subjects, an increase of 50 per cent. The ratio of the area of exposure of capillary blood to each square millimeter of skin was found to be three times greater than in normal skin. These data indicate that capillary distention is more important than numerical increase in the number of loops, this is further emphasized by the effect of blood reduction on the size and number of capillary loops.

CARDIAC OUTPUT IN PNEUMONIA IN THE DOG

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(Received for publication April 5, 1926)

INTRODUCTION

The condition of the circulation of patients suffering from pneumonia is always of definite concern to the physician. An attempt is commonly made to discover the onset of signs of cardiac failure, and to evaluate the degree of failure when these signs appear, as circulatory failure is an important factor leading to a fatal outcome in many cases of pneumonia. The mechanism of cardiac failure in pneumonia, however, is not well understood, and no satisfactory explanation has been offered for the relation of disturbance of the circulation to the pulmonary lesions or to the effects of the organisms responsible for the disease. Doubt has been cast on the assumption that failure of the heart is the result of the direct action of the toxins of the infecting organism on the myocardium. Newburgh and Porter (1) found that strips of muscle from the cardiac ventricles of dogs dying of pneumonia contracted as well as similar strips from normal dogs when both were perfused with normal blood. When "pneumonic blood" was used as a perfusate the heart muscle of pneumonic dogs contracted better than the muscle of normal dogs. These results led Newburgh and Porter to assume the existence in pneumonic blood of a toxin to which the animals dying of the disease had become relatively immune.

From a review of the literature and from their own studies Cohn and Jamieson (2) concluded that significant alterations in the heart muscle are not generally found in pneumonia.

The question of the effect of the pulmonary lesion on the circulation has received but scant attention. On general principles it is evident that if the functional efficiency of the lungs is lowered so that the blood is supplied with less than its normal quota of oxygen in its

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2 *Post-operative pneumonia* The cardiac output was studied in nine dogs in which pneumonia developed following various operations—most frequently after thoracotomies

3 *Induced pneumonia* Intrabronchial injection of pneumococci or Friedlander bacilli was done in thirteen dogs

In the animals of the first two groups control studies were not available, while in the third group the cardiac output was measured before and after the injection of organisms. Difficulty was encountered in the study of the third group in the regulation of the dosage of the organisms injected, as dosage was either so large as to kill the animal in a few hours, or so small as to produce no appreciable effect. Of thirteen dogs inoculated only six lived long enough to develop clinical evidence of pneumonia. When the dose was small the animals appeared to recover very rapidly. With large doses death occurred apparently from an overwhelming infection, without the characteristic picture of pneumonia. From our results we conclude that only one of the dogs with induced pneumonia had the disease long enough to develop a picture simulating the more chronic pneumonia of the other groups.

Most of the animals received morphine 0.006 gram per kilo, 30 to 60 minutes before each determination. Several determinations were done without morphine and the results were similar to those found in the dogs receiving the drug. This indicates that the effect of morphine on cardiac output was negligible in these experiments. Marshall (5) has found that morphine had no appreciable effect on cardiac output of the normal dog, and Blalock, Harrison, and Wilson (6) using morphinized dogs found results for blood flow only slightly higher than the result of Marshall on unanesthetized trained dogs.

The oxygen consumption was determined by means of a Benedict spirometer with the graphic recording device. In earlier experiments a rubber bladder was made air tight over the animal's head in order to obtain an accurate measurement of the respiration. In later experiments a paraffined plaster of paris mask was fitted to the dog's nose and made air tight with plasticine.

Venous blood was obtained by puncture of the right ventricle, arterial blood from the left ventricle or the femoral artery. Two samples of blood were usually drawn from the right ventricle in order to guard against errors arising from the possible mixture of arterial blood with the venous blood. The usual precautions against contact with air were followed. The arterial and venous oxygen contents were determined with the Van Slyke (7) long bore, constant pressure apparatus in the first experiments, the later determinations were made with the Van Slyke-Neill (8) constant volume apparatus. Oxygen capacity was also determined in some instances.

The cardiac output per minute was calculated according to the Fick formula—

$$\frac{\text{cc. oxygen consumed per minute}}{\text{cc. oxygen taken up by 1 cc. of blood in passing through the lungs}} = \text{cc. blood passing through lungs per minute}$$

passage through the pulmonary circulation, an increased blood flow is necessary to meet the oxygen requirements of the body. For this reason the task of the heart is increased when pulmonary consolidation interferes with the gas exchange in the lungs. The augmented demand of the body for oxygen which accompanies fever adds a further burden on the heart, so that in pneumonia the demands on the circulation may be especially heavy, and may at times surpass the ability of the circulation to respond. Then signs of circulatory failure appear.

Few investigations of the circulation in pneumonia from the point of view of its respiratory functions have appeared. Stadie (3) studied the arterial and venous oxygen contents of the blood of patients with pneumonia. He stated "The oxygen consumption, i.e., difference between arterial and venous contents was within normal limits, indicating that the cardiac output was not diminished." His figures show a low rather than a high arterio-venous difference. Assuming an increased rate of tissue oxidation, such as is usually found in fevers, one is led to believe from Stadie's data that cardiac output was probably actually increased.

Leegaard (4) recently published preliminary observations on cardiac output in rabbits after pneumonia had been induced by the intratracheal injection of pneumococci. He found a 50 to 100 per cent increase in the minute output of the heart.

Our interest in the problem followed the accidental finding of a high cardiac output in a supposedly normal dog, which at autopsy was found to have extensive broncho-pneumonia. Since then eighty-seven determinations of blood flow have been made on twenty-four additional dogs. In this paper the various factors causing variations in the output of the heart in pneumonia are analyzed and discussed. Observations regarding other factors bearing on the problem of variations in the cardiac output will be considered in subsequent papers.

METHODS

Three different types of pneumonia have been studied.

1 *Spontaneous pneumonia* Measurements of the cardiac output were made on a number of stock animals showing signs of severe distemper, emaciation, and fever. After two or more determinations the animals were sacrificed. Three of the group were found at necropsy to have pneumonia.

TABLE 1
Pneumonia following operation
 Animal I2 Weight varied 4.6 to 5.9 kilos

Date	Pulse rate	Temperature (Fahrenheit)	Arterial oxygen	Venous oxygen	Oxygen capacity	Saturation	pH	Oxygen consumption per minute	Cardiac output per minute	Output per minute per kilo	Output per beat
	<i>per minute</i>		<i>volumes per cent</i>	<i>volumes per cent</i>	<i>volumes per cent</i>	<i>per cent</i>		<i>cc.</i>	<i>cc.</i>	<i>cc.</i>	<i>cc.</i>
5-10	68	100.1	15.1	12.7	17.4	86.8		36.4	1,517	330	22.3
5-11	56	100.5	14.4	10.5	17.1	84.2		52.7	1,351	294	24.0
5-23	54	99.8	15.4	11.4			7.28	38.2	1,151	250	21.3
5-26	62	98.0	14.6	10.8				37.6	989	219	16.0
6-17	64		19.6	13.7				43.7	741	148	11.6
10-3	60	99.0	22.02	17.31				44.7	949	160	15.8
10-22	78	102.0	18.27	11.49				56.4	725	123	9.0

(Figures in italics denote determinations done when animal was apparently normal, other figures denote determinations when pneumonia was thought to be present.)

Protocol April 21, 1925, pericardotomy performed May 7, dog looks sick. May 11, chest X-rayed, thought to show pneumonia. June 23, dog seems very much improved, gaining weight. October 10, looks well. October 20, listless and febrile, without evidence of distemper or pneumonia. (Morphine 0.04 gram given before each determination.)

TABLE 2
Pneumonia following operation
 Animal C8 Weight varied 8.0 to 7.7 kilos

Date	Pulse rate	Arterial oxygen	Venous oxygen	Oxygen capacity	Saturation	pH	Oxygen consumption per minute	Cardiac output per minute	Output per minute per kilo	Output per beat
	<i>per minute</i>	<i>volumes per cent</i>	<i>volumes per cent</i>	<i>volumes per cent</i>	<i>per cent</i>		<i>cc.</i>	<i>cc.</i>	<i>cc.</i>	<i>cc.</i>
12-9-24	45	20.4	15.4	21.9	93.1		48.1	962	120	21.4
1-4-25	56	17.8	15.6	20.2	88.1	7.33	48.1	2,187	284	39.0

(Figures in italics denote determinations done when animal was apparently normal, other figures denote determinations when pneumonia was thought to be present.)

Protocol December 9, 1924, tracheotomy. December 16, dog has distemper. January 4, 1925, distemper seems worse, morphine 0.06 gram before determination. Extensive bilateral pneumonia found at autopsy.

The hydrogen ion concentration of the blood was determined in a few instances, Hawkins (9) modification of Cullen's (10) colorimetric method being used

When the dogs struggled, or when a reasonable suspicion was entertained that either venous or arterial blood was "mixed" the experiment was discarded

Duplicate five-, or ten-minute periods were used in determining the oxygen consumption. Check determinations were made of the oxygen content of the blood. Single determinations of oxygen capacity were considered sufficiently reliable

Most of the animals died spontaneously. A few were killed either intentionally or by the production of hemopericardium. The gross anatomical findings were confirmed by microscopic examination of the tissues. The output of the heart per minute is for the sake of comparison divided by the kilograms of body weight of each animal. The output per minute is also divided by the number of heart beats per minute in order to obtain the average cardiac output per beat

RESULTS

The details of the results of each experiment are given in the accompanying tables

The results of the experiments on dogs with spontaneous pneumonia and on dogs developing post-operative pneumonia are similar and are therefore considered together. In the eleven animals constituting these two series the cardiac output varied between 200 and 400 cc of blood per minute per kilo of body weight, and averaged 274 cc. These figures appear in sharp contrast to those obtained from normal dogs, in which the cardiac output has been found to be from 100 to 200 cc per minute per kilo. In a series of 18 normal dogs the cardiac output averaged 167 cc per minute per kilo. Thus it is evident that the dogs with spontaneous and post-operative pneumonia had a striking increase in the output of the heart per minute.

The series of animals in which pneumonia was induced by the intrabronchial injections of bacteria gave results regarding the output of the heart which are quite different from those obtained in the other two groups (tables 11 to 14). In this series of six dogs, with one exception, no significant change in the cardiac output was observed after pneumonia was induced when compared with the measurements made before the organisms were inoculated into the bronchi. In one animal a definite increase in the output of the heart was observed. The average results of the entire series are as follows

	Cardiac output per minute per kilo
	cc.
1 Before inoculation (eleven determinations)	170
2 After inoculation (thirteen determinations)	199
3 After inoculation (twelve determinations)	179

TABLE 5
Pneumonia following operation

Date	Pulse rate	Arterial oxygen	Venous oxygen	Oxygen consumption per minute	Cardiac output per minute	Output per minute per kilo	Output per beat
Animal 5 Weight 6 kilos							
	<i>per minute</i>	<i>volumes per cent</i>	<i>volumes per cent</i>	<i>cc</i>	<i>cc</i>	<i>cc</i>	<i>cc</i>
4-5	76	19.2	16.7	56.0	2,240	373	29.4
Animal 6 Weight 5.3 kilos							
4-12	56	16.5	11.1	41.2	763	143	13.6

Protocol Animal No 5 March 7, 1925, pericardotomy April 2, animal appears sick. April 5, very weak, unable to stand April 6, dead Lungs showed extensive bilateral broncho-pneumonia

Protocol Animal No 6 March 14, 1925, pericardotomy April 8, animal appears quite sick. April 12, seems improved April 16, animal dead. Many pericardial adhesions and a few scattered patches of broncho-pneumonia found at autopsy (Given 0.04 gram of morphine before determinations)

TABLE 6
Pneumonia following operation
Animal V31 Weight 6.7 kilos

Date	Pulse rate	Temperature (Fahrenheit)	Arterial oxygen	Venous oxygen	Oxygen capacity	Saturation	Oxygen consumption per minute	Cardiac output per minute	Output per minute per kilo	Output per beat
	<i>per minute</i>		<i>volumes per cent</i>	<i>volumes per cent</i>	<i>volumes per cent</i>	<i>per cent</i>	<i>cc.</i>	<i>cc.</i>	<i>cc.</i>	<i>cc.</i>
2-20 1 p.m.	48	97.0	20.68	14.99	21.5	96.2	45.6	801	119	16.6
2-20 3 p.m.	52	97.4	18.49	12.58	20.23	91.4	46.9	808	120	15.5
2-22 9 a.m.	62	100.2	12.36	9.12	16.44	75.2	57.6	1,777	278	28.6
2-22 11 a.m.	56		12.65	8.10	16.12	78.5	56.9	1,248	192	22.0

(Figures in italics denote determinations done when animal was apparently normal, other figures denote determinations when pneumonia was thought to be present.)

Protocol February 20, 1925, tracheotomy performed. Two control determinations on this date February 22, appears weak, neck wound infected February 23, animal killed Complete consolidation left upper lobe and patchy consolidation elsewhere found at autopsy

If the one experiment in which an increased cardiac output following inoculation is omitted from the series, the average results indicate that no increase in the minute output of the heart took place after inoculation

TABLE 3
Pneumonia following operation
Animal I3 Weight 5.1 kilos

Date	Pulse rate	Temperature (Fahrenheit)	Arterial oxygen	Venous oxygen	Oxygen consumption per minute	Cardiac output per minute	Output per minute per kilo	Output per beat
	<i>per minute</i>		<i>volumes per cent</i>	<i>volumes per cent</i>	<i>cc</i>	<i>cc</i>	<i>cc.</i>	<i>cc</i>
5-10	98	104.8	14.5	7.1	50.3	679	133	7.1
5-11	122	102.0	13.2	7.8	48.9	906	177	7.4

Protocol April 25, 1925, pericardotomy During the following week animal was listless and refused to eat May 11, seems much sicker May 12, extensive bronchopneumonia and adhesive pericarditis found at autopsy

TABLE 4
Pneumonia following operation
Animal I4 Weight varied 6.4 to 5.6 kilos

Date	Pulse rate	Temperature (Fahrenheit)	Arterial oxygen	Venous oxygen	Oxygen capacity	Saturation	pH	Oxygen consumption per minute	Cardiac output per minute	Output per minute per kilo	Output per beat
	<i>per minute</i>		<i>volumes per cent</i>	<i>volumes per cent</i>	<i>volumes per cent</i>	<i>per cent</i>		<i>cc</i>	<i>cc</i>	<i>cc.</i>	<i>cc.</i>
5-21	55	103.6	16.4	12.7	20.2	81.2		74.0	2,000	313	36.3
5-25	82	102.2	7.7	3.8	12.3	62.6		63.0	1,620	270	19.7
5-27	63	101.5	8.0	3.0	11.8	67.8	7.31	60.1	1,205	215	19.1

Protocol April 28, 1925, nerve suture operation performed May 12, distemper noticed Animal appeared quite sick and began to lose weight May 21, looks very sick May 25, continues to appear sick X-ray shows extensive consolidation on both sides May 27, moribund May 28, found dead Complete consolidation of left lung and patchy consolidation of right lung found at autopsy (Morphine 0.04 gram given before each determination)

The rectal temperature was slightly elevated in most, but not all, of the dogs with pneumonia The pulse rate was sometimes moderately increased, but pulse rates are of little significance in morphinized

ished. The emaciation was not of as great a degree as was the increase in cardiac output and a relatively small fraction of the increased blood flow per kilo can be attributed to undernutrition.

TABLE 9
Spontaneous pneumonia
Animal V6 Weight varied 8.4 to 7.8 kilos

Date	Pulse rate	Temperature (Fahrenheit)	Arterial oxygen	Venous oxygen	Oxygen consumption per minute	Cardiac output per minute	Output per minute per kilo	Output per beat
	per minute		volumes per cent	volumes per cent	cc.	cc.	cc.	cc.
10-12	109	102.2	11.37	6.3	104.0	2,051	244	18.8
10-13	110	102.0	11.59	5.94	91.5	1,622	196	14.7
10-18	114	102.0	11.76	6.4	94.5	1,763	218	15.4
10-24	118	102.6	12.33	7.76	104.4	2,276	284	18.4
11-2	115	101.8	12.38	7.53	95.7	1,973	253	17.1

Protocol: October 2, 1925, distemper first noticed. October 12, X-ray thought to show bilateral patchy pneumonia. October 18, animal appears slightly improved, less discharge from nose. October 24, X-ray shows clearing of apical shadows.

TABLE 10
Spontaneous pneumonia
Animal V12 Weight 11.4 kilos

Date	Pulse rate	Temperature (Fahrenheit)	Arterial oxygen	Venous oxygen	Oxygen capacity	Saturation	Oxygen consumption per minute	Cardiac output per minute	Output per minute per kilo	Output per beat
	per minute		volumes per cent	volumes per cent	volumes per cent	per cent	cc.	cc.	cc.	cc.
11-25	68	100	14.55	11.16	16.10	90	79.1	2,363	207	34.7
11-27			14.60	11.28			81.4	2,420	212	

Protocol: November 25, 1925, animal looks healthy. Temperature slightly elevated, blood flow very high. November 27, animal killed after experiment completed. Autopsy showed chronic broncho-pneumonia of nodular type. Very extensive pneumonia found on microscopic study.

Roentgenograms were taken in some instances and compared with plates from a normal group. The radiological findings were considerably less striking than the post mortem evidences of pneumonia.

TABLE 7
Spontaneous pneumonia
 Animal M2 Weight varied 6.2 to 6 kilos

Date	Pulse rate	Temperature (Fahrenheit)	Arterial oxygen	Venous oxygen	Oxygen capacity	Saturation	Oxygen consumption per minute	Cardiac output per minute	Output per minute per kilo	Output per beat
	per minute		volumes per cent	volumes per cent	volumes per cent	per cent	cc	cc	cc	cc
5-21		101.2	11.9	8.5	13.3	89.5	80.3	2,362	381	
5-23	98	104.6	4.4	2.6	10.1	43.6	85.8	4,760	768	48.5

Protocol May 7, 1925, distemper first noticed May 21, dog has lost a great deal of weight Profuse discharge from nose and eyes First determination on blood flow May 23, unable to stand Second determination May 24, found dead Complete consolidation right lower lobe and extensive patches of pneumonia throughout the rest of the lungs were found at autopsy (Morphine 0.03 gram given before each determination)

TABLE 8
Spontaneous pneumonia
 Animal V7 Weight varied 7.8 to 6.6 kilos

Date	Pulse rate	Temperature (Fahrenheit)	Arterial oxygen	Venous oxygen	Oxygen capacity	Saturation	pH	Oxygen consumption per minute	Cardiac output per minute	Output per minute per kilo	Output per beat
	per minute		volumes per cent	volumes per cent	volumes per cent	per cent		cc	cc	cc	cc
10-27	126	101	8.93	4.80	10.74	83.1		112.8	2,731	350	21.6
10-28	148	101	7.98	4.51	10.82	73.7		93.4	2,697	346	18.2
10-31	101	99	7.09	3.29	9.91	71.5	7.22	73.4	1,932	293	19.1

Protocol September 30, 1925, distemper first noticed Dog slightly emaciated October 27, profuse discharge from nose and eyes Animal very weak October 31, apparently improved November 1, found dead At autopsy, almost complete consolidation of right lower lobe and slight consolidation of left lower lobe were found (No morphine used)

dogs, as the drug causes a marked slowing of the heart Anaemia was found in most of the animals but the degree was variable Some of the dogs were much emaciated For this reason the blood flows per kilo are higher than would be the case had they been well nour-

Hydrogen ion concentrations were determined in six animals. Of these, four had extensive pneumonia with a high cardiac output

TABLE 13
Induced pneumonia
Animal V4 Weight varied 6.4 to 5.7 kilos

Date	Pulse rate	Temperature (Fahrenheit)	Arterial oxygen	Venous oxygen	Oxygen consumption per minute	Cardiac output per minute	Output per minute per kilo	Output per beat
	<i>per minute</i>		<i>volumes per cent</i>	<i>volumes per cent</i>	<i>cc</i>	<i>cc</i>	<i>cc</i>	<i>cc</i>
12-4	58	100.0	15.74	9.45	54.0	846	132	14.5
12-5	88	98.0	16.96	10.65	58.9	933	155	10.6
12-7	64	99.0	16.49	10.29	57.0	921	161	14.3
12-10	100	102.2	13.51	7.91	60.4	1,079	189	10.7

(Figures in italics denote determinations done when animal was apparently normal, other figures denote determinations when pneumonia was thought to be present.)

Protocol December 4, 1925, control studies. December 5, second control studies. After determination given 10 cc. of 19-hour growth Type III pneumococcus. December 7, does not look sick. December 10, somewhat dyspneic. Dog died of hemopericardium shortly after last puncture was made. At autopsy, the lung bases seemed heavy and the upper lobes normal. Microscopic sections show much congestion, extensive oedema, and fairly extensive leucocytic exudation. (Morphine 0.04 gram given before each determination.)

TABLE 14
Induced pneumonia
Animal V15 Weight 10.2 kilos

Date	Pulse rate	Temperature (Fahrenheit)	Arterial oxygen	Venous oxygen	Oxygen consumption per minute	Cardiac output per minute	Output per minute per kilo	Output per beat
	<i>per minute</i>		<i>volumes per cent</i>	<i>volumes per cent</i>	<i>cc</i>	<i>cc</i>	<i>cc</i>	<i>cc</i>
12-14	60	97.0	21.56	17.1	85.9	1,928	189	32.1
12-15	84	100.2	18.06	13.86	76.4	1,819	178	21.6

(Figures in italics denote determinations done when animal was apparently normal, other figures denote determinations when pneumonia was thought to be present.)

Protocol December 14, control studies. Forty cubic centimeter of 18-hour growth Type III pneumococcus given after determination. December 15, listless and very weak. December 16, found dead. Autopsy revealed extensive consolidation of all lobes, confirmed by microscopic sections.

and two a less extensive pneumonia with normal blood flow. The pH values all fell within the range (7.20-7.35) found as normal for morphinized dogs by Harrison, Wilson and Blalock (11).

Induced pneumonia

Animal M1 Weight varied 13.2 to 11.3 kilos

Date	Pulse rate	Temperature (Fahrenheit)	Arterial oxygen	Venous oxygen	Oxygen capacity	Saturation	Oxygen consumption per minute	Cardiac output per minute	Output per minute per kilo	Output per beat
	<i>per minute</i>		<i>volumes per cent</i>	<i>volumes per cent</i>	<i>volumes per cent</i>	<i>per cent</i>	<i>cc</i>	<i>cc.</i>	<i>cc.</i>	<i>cc</i>
5-26	76	99.0	23.7	18.9	24.1	98.3	94.0	1,920	153	25.7
5-30	82	102.0	22.0	17.3	22.0	100.0	105.9	2,253	173	27.4
6-7	79	103.2	19.8	14.4	20.5	96.6	120.4	2,230	186	28.2
6-18	63	98.8	19.3	14.4			86.3	1,761	140	27.9
6-20	130	101.6	12.1	8.2	17.9	67.6	97.3	2,497	189	19.2

(Figures in italics denote determinations done when animal was apparently normal, other figures denote determinations when pneumonia was thought to be present.)

Protocol May 26, 1925, healthy dog Control determination May 30, second control determination June 6, inoculated with 40 cc of 24-hour growth Type II pneumococcus June 7, temperature elevated, restless, looks well June 18, apparently recovered Inoculated again after determination June 19, looks sick dyspneic, cyanotic, unable to stand June 20, somewhat improved, given 20 cc of 30 hour growth of Friedlander bacilli June 21, condition unchanged June 22, killed Complete consolidation right middle and lower lobes, few scattered patches elsewhere Findings confirmed by microscopic sections

TABLE 12

Induced pneumonia

Animal M5 Weight varied 12 to 10.6 kilos

Date	Pulse rate	Temperature (Fahrenheit)	Arterial oxygen	Venous oxygen	Oxygen capacity	Saturation	Oxygen consumption per minute	Cardiac output per minute	Output per minute per kilo	Output per beat
	<i>per minute</i>		<i>volumes per cent</i>	<i>volumes per cent</i>	<i>volumes per cent</i>	<i>per cent</i>	<i>cc</i>	<i>cc</i>	<i>cc.</i>	<i>cc</i>
5-30	63	100.2	16.5	11.9	17.8	92.7	114.1	2,480	207	39.3
6-2	85	104.5	16.7	11.5	17.9	93.3	117.0	2,250	201	26.4
6-4	82	102.0	13.7	7.9	16.0	85.6	114.4	1,972	182	24.0
6-5	136	102.4	13.6	8.0	14.8	91.9	109.4	1,954	183	14.3
6-8	108	102.5	3.8	2.0	5.1	74.5	88.5	4,917	464	45.5

(Figures in italics denote determinations done when animal was apparently normal, other figures denote determinations when pneumonia was thought to be present.)

Protocol May 30, 1925, control studies June 1, given 28 cc of 24-hour growth Type I pneumococcus June 2, looks healthy, not cyanotic, high fever June 3, looks very sick June 4, condition unchanged June 8, animal is very weak Autopsy performed after experiment was completed showed the lungs to be normal in the gross except for slight consolidation of both lower lobes Microscopic sections show fairly extensive early broncho-pneumonia

Hastings, Neill, Morgan and Binger (12) found normal values for pH in pneumonic patients, whereas Leake, Vickers and Brown (13) found slight acidosis in the experimental pneumonia of dogs

The animals developed a marked anaemia in many instances. The cause of this is not clear. Leake, Vickers, and Brown (13) found that in experimental pneumonia of dogs a great loss of circulating red blood cells occurred without compensatory regeneration. From our studies it is not possible to be sure that the diminution of oxygen was dependent entirely on loss of red blood corpuscles as counts were not made. It is possible that methemoglobin may have been present. If so the amount of methemoglobin was not significant, as no change was noted in the color of the aerated blood.

DISCUSSION

The results of the experiments here reported are discussed from two points of view

1 *The mechanism responsible for the increased output of the heart in pneumonia*. Our present knowledge of the controlling factors of the circulation is comparable to our knowledge of the regulation of respiration before the classical observations of Zuntz (14) and Haldane and Priestley (15), demonstrating the importance of oxygen and carbon dioxide. A rational conception of the regulation of the cardiac output is of prime importance to clinicians as well as to physiologists. Therefore an attempt has been made to analyze the various factors which may have been responsible for the increased cardiac output in pneumonia. In an investigation now in progress these factors are being studied separately in an endeavor to ascertain their relationship to the circulation under less complicated conditions.

In pneumonia there are at least five abnormal conditions which may lead to alterations of the output of the heart. These are

- 1 The effect of toxins produced by the infecting organisms on the tissues, producing fundamental disturbances of function
- 2 Acidosis (if present)
- 3 Increased metabolic rate
- 4 Anaemia
- 5 Lack of oxygen in the blood leaving the lungs

TABLE 15
Induced Pneumonia
Animal V2 Weight varied 13.6 to 11.4 kilos

Date	Pulse rate	Temperature (Fahrenheit)	Arterial oxygen	Venous oxygen	Oxygen capacity	Saturation	pH	Oxygen consumption per minute	Cardiac output per minute	Output per minute per kilo	Output per beat
	<i>per minute</i>		<i>volumes per cent</i>	<i>volumes per cent</i>	<i>volumes per cent</i>	<i>per cent</i>		<i>cc</i>	<i>cc</i>	<i>cc</i>	<i>cc</i>
10-8	67		17.71	13.43	19.21	92.2		85.2	1,990	146	29.7
10-19	71	98.0	17.70	13.25				98.6	2,216	181	31.2
10-22		100.0	17.45	13.57				82.6	2,123	174	
10-29	113	101.2	12.82	7.77	15.04	85.2	7.27	87.5	1,733	154	15.3

(Figures in italics denote determinations done when animal was apparently normal, other figures denote determinations when pneumonia was thought to be present.)

Protocol October 8, 19, and 22, control studies October 23, X-ray shows normal lungs October 26, 35 cc of 10-hour growth Type I pneumococcus October 27, slightly dyspneic, temperature 102 October 29, weak, losing weight Killed after determination The left upper lobe was solid Patches of broncho-pneumonia throughout rest of lungs Microscopic sections showed extensive pneumonia

TABLE 16
Induced pneumonia
Animal V5 Weight varied 5.9 to 5.6 kilos

Date	Pulse rate	Temperature (Fahrenheit)	Arterial oxygen	Venous oxygen	Oxygen capacity	Saturation	Oxygen consumption per minute	Cardiac output per minute	Output per minute per kilo	Output per beat
	<i>per minute</i>		<i>volumes per cent</i>	<i>volumes per cent</i>	<i>volumes per cent</i>	<i>per cent</i>	<i>cc</i>	<i>cc</i>	<i>cc</i>	<i>cc</i>
10-25	68	98.0	16.3	11.22			56.7	1,116	189	16.4
10-26	92	97.6	14.24	9.41	16.01	88.9	62.6	1,296	219	14.0
10-29	110	102.0	15.01	8.05			74.4	1,069	191	9.7
11-1	82	102.0	13.78	6.40	15.52	88.8	72.8	986	176	12.0

(Figures in italics denote determinations done when animal was apparently normal, other figures denote determinations when pneumonia was thought to be present.)

Protocol October 25, 1925, control study X-ray showed normal lungs October 26, second control study After determination given 20 cc of 10-hour growth Type I pneumococcus October 27, somewhat weak, slightly dyspneic, temperature 103 October 29, condition unchanged November 1, improved, walking around, studies without morphine Killed after determination Autopsy showed few patches broncho-pneumonia both lower lobes (Morphine, $\frac{3}{4}$ grain, given before each determination with the exception of the one on November 1.)

Satisfactory normal standards are not available for determining whether the basal metabolic rate of a dog is increased. Most of the dogs with pneumonia had some elevation of body temperature and an increased heat production may be assumed. The oxygen consumption per minute per kilo varied from 6 to 14 cc, and was in general in inverse ratio to the weight of the dog. In figure 2 a correlation is shown between the number of cubic centimeters of

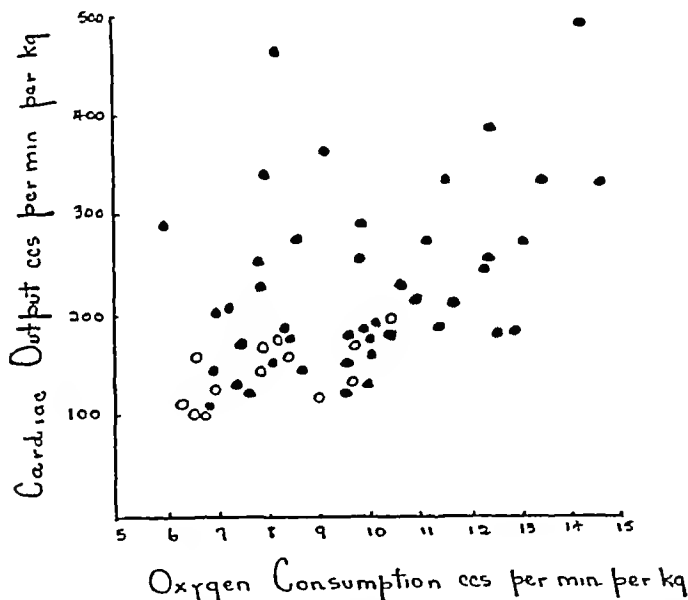


FIG 2 The cardiac output per minute per kilo is charted against the oxygen consumed per minute per kilo. There is a general tendency for the cardiac output to be higher in the animals with a higher oxygen consumption, but this is not striking.

oxygen consumed per minute per kilogram and the cardiac output per minute per kilo. Although the higher values for the cardiac output coincide in a rough way with the higher values for oxygen consumption, the relationship is not striking.

A similar chart (fig 3) depicts the correlation between the cardiac output and hemoglobin content (the values for hemoglobin were calculated from the oxygen capacities on the assumption that 20

The effect of toxins cannot be studied quantitatively with present methods, and so this possibility has been neglected in our further consideration

Since Leake, Brown and Vickers found acidosis in dogs with pneumonia, and since Harrison, Wilson and Blalock found that acidosis produces an increase in the cardiac output of morphinized dogs, at-

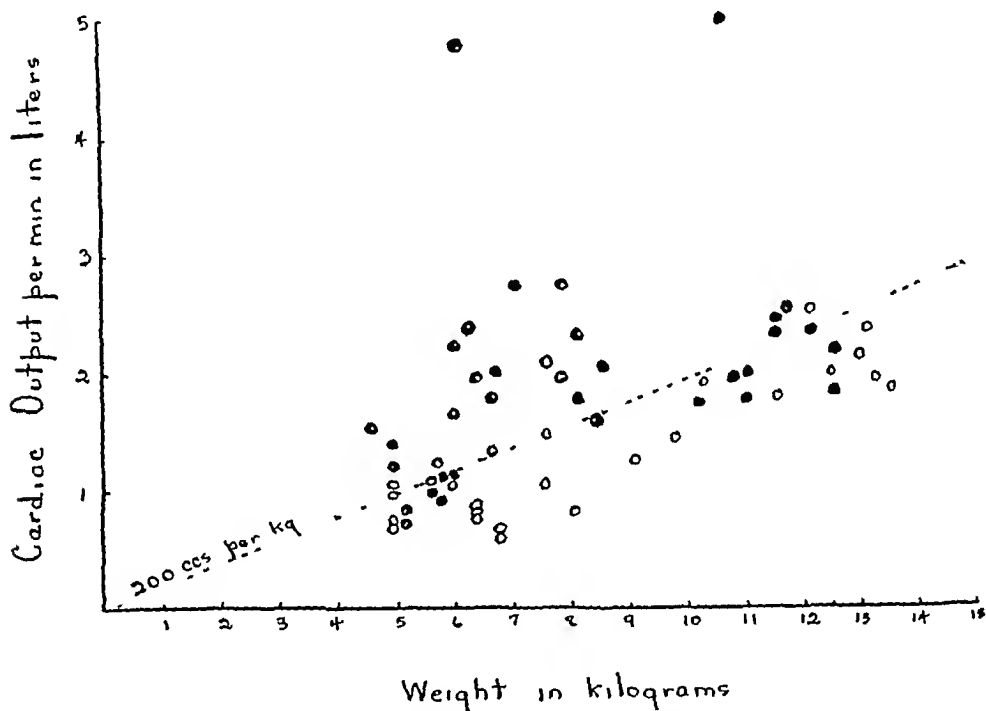


FIG 1 The total cardiac output per minute is charted against the weights. The diagonal dotted line represents 200 cc per minute per kilo of body weight. In this and the following figure the white circles denote normal dogs, the black circles—dogs with pneumonia. Nearly all the normal animals have a cardiac output of less than 200 cc per kilo per minute, whereas most of the dogs with pneumonia have a higher output.

tention was first directed to this factor. However, as has been stated above, the pH value of the blood of the animals with pneumonia was found to fall within the same range as the blood pH value of normal dogs receiving similar doses of morphine. The pH decline necessary to produce a marked increase in cardiac output is greater than the degree of this morphine acidosis, as Harrison, Wilson and Blalock have shown.

2 *The clinical significance of an increased cardiac output in pneumonia* If one makes the assumption that the human heart responds to the pneumonic process in the same way as the heart of the rabbit (Leegaard) or dog—an assumption which seems likely but remains to be proved—certain probable deductions can be drawn

a When the infection is overwhelming and death comes in one to three days, there is little or no change in cardiac output When the

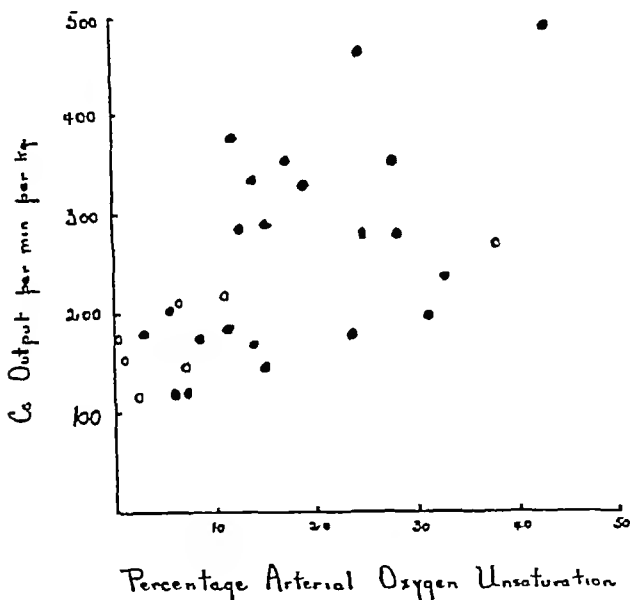


FIG 4 The cardiac output per minute per kilo is charted against the percentage of arterial unsaturation—or degree of anoxemia With high degrees of arterial unsaturation the cardiac output is greatly increased

infection is mild there is relatively little change The severe but not immediately overwhelming infection of several days standing apparently causes the greatest increase in blood flow In these instances the cardiac output seems to stand in the same relation to the severity of the infection as does the leucocyte count

b The assumption that death occurs in pneumonia from cardiac failure seems to be true in one sense and false in another The older conceptions of a heart muscle injured by toxin contracting more

volumes per cent capacity equal 100 per cent hemoglobin). From this figure it appears that the relationship of increased output of the heart to anaemia is fairly definite, although caution is necessary in interpreting results when other complicating factors are present

Cardiac output in relation to anoxemia is demonstrated in figure 4. Here the correlation is more suggestive than with the functions pre-

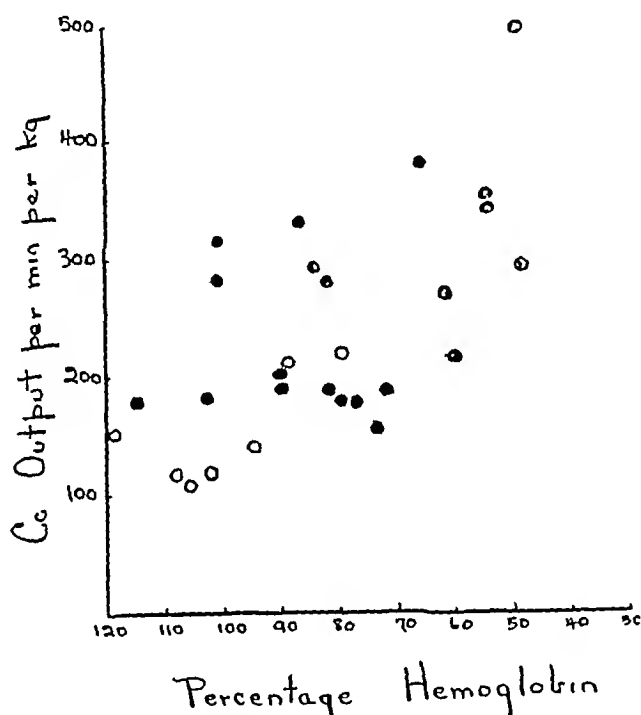


FIG 3 The cardiac output per minute per kilo is charted against the percentage hemoglobin, which was calculated from the oxygen capacity on the assumption that 20 volumes per cent oxygen capacity equals 100 per cent hemoglobin. The reduction of hemoglobin is strikingly correlated with an increase in cardiac output.

viously plotted, but one is scarcely justified in drawing any definite conclusions under the complicated conditions of the experiments.

These results indicate that increased metabolic rate, anaemia of severe degree, and anoxemia may be of importance in regulating the blood flow. In order to evaluate each of these factors it is necessary that they be studied singly. This is being done at the present time and the results will be published at a later date.

phoid fever and acute miliary tuberculosis, comparable as to degree of "toxemia "

The relationship of anoxemia to the increased cardiac output has been discussed above, and severe anoxemia appears to be correlated with increased output of the heart. This affords a rational basis for the administration of oxygen in pneumonia. Increasing the alveolar oxygen tension should diminish the necessity for an increased cardiac output. If the heart of man reacts to pneumonia as does the heart of the dog, oxygen should have a very beneficial sedative action in those cases where cyanosis is marked.

In comparing the findings in experimental pneumonia with clinical pneumonia it is necessary to remember that the latter is frequently encountered in individuals with diseased hearts, whereas the experimental animals had normal hearts, so far as we could judge. It is possible that the pathological heart cannot increase its output to any considerable degree and this may account for the well known fact that pneumonia is more fatal in individuals suffering from cardiac diseases.

The value of "cardiac stimulants" remains an open question. Caffeine and camphor are commonly used. Aside from their value as respiratory stimulants, our purpose in giving them is to increase the output of the heart. If these drugs do not have this effect it is useless to give them. If they do have this effect, their value is questionable and possibly their use is harmful. Our knowledge of the circulation is too vague to permit an answer to the question of the advisability of increasing the work of an organ already working at or near its maximum level. Clinically one gains the impression that these drugs are of little value except in those patients who develop Cheyne-Stokes breathing. However, the "cardiac stimulants" are usually not given until the patient is well nigh moribund and therefore such an impression may not be correct. Further experiments and better methods for study of patients are needed before caffeine and camphor can be evaluated.

Digitalis is widely used in pneumonia. Cohn and Jamieson demonstrated that this disease does not alter the effect of the drug on the heart.

feebly seems incorrect. From the results of others, quoted above, and of this work, it is more logical to think of a heart doing more than the normal amount of work and failing eventually because of the increased load.

Of practical importance is the question as to whether the increased blood flow in pneumonia is a compensatory and hence necessary mechanism, or whether this increase is to be regarded as being *per se* a dangerous manifestation of the disease and leading eventually to failure of the circulation. It appears probable that both conceptions are correct. Blalock, Harrison and Wilson have shown experimentally that diminishing the minute ventilation by partially obstructing the respiration causes a great increase in the output of the heart. From their experiments they concluded that the circulation constitutes the second line of defense against failure of tissue metabolism and that the heart by increased output can compensate for an inefficient respiratory mechanism. In their experiments the CO_2 elimination was markedly affected while relatively little change in the oxygen absorption was found. In the experiments on pneumonia reported in this paper the reverse is true, but the circulatory response is similar. We believe that either a rise of alveolar CO_2 tension or a fall in alveolar oxygen tension, if of sufficient magnitude, will cause an increased cardiac output. The advantage to the body is obvious. The amount of oxygen taken up, or of CO_2 given off per 1 cc of blood in the lungs depends on the difference between the pressure of these gases in the alveoli and in the venous blood. If the alveolar CO_2 tension rises, each unit of blood can give off less CO_2 . If the alveolar oxygen tension falls (beyond a certain level) each unit of blood can take up less oxygen. Hence, in either case, if the total exchange of gas is to remain constant more units of blood must pass through the lungs. The increased blood flow must be regarded therefore as a compensatory mechanism and an advantage, but obtained at the risk of possible cardiac failure. If the heart can carry the increased load until the infection is overcome, recovery may be expected, if the strain on the heart causes it to weaken, death results.

It is believed that these conceptions explain why circulatory failure occurs more rapidly in pneumonia than in other diseases, such as ty-

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Levy (16) has shown that digitalis diminishes the size of the heart shadow in pneumonia. In some work now in progress Leonard and Harrison have found that digitalis in therapeutic doses diminishes the output of the dog's heart. If we may believe that the dog's heart and human heart are comparable, one is led to assume that most patients with pneumonia need a cardiac sedative rather than a cardiac stimulant. Although the increased blood flow is probably compensatory, the heart must be protected against compensating to the extent of becoming weakened by the strain.

SUMMARY

The output of the heart of the dog has been studied by the Fick method. In pneumonia the cardiac output is usually increased. When the infection is of short duration and either mild or overwhelming, the increase is less than in the severe, well developed pneumonia of longer duration. The increase in cardiac output appears to bear little relation to changes in oxygen consumption, but appears to depend to some extent on anaemia and to a greater extent on anoxaemia. On the assumption that the cardiac output is also increased in pneumonia in man, heart failure is to be ascribed to the increased strain on the circulation rather than to weakness of the myocardium. From this viewpoint oxygen and digitalis appear to be definitely indicated, whereas the value of caffeine and camphor is questionable.

It is a pleasure to express our appreciation to Dr G. Canby Robinson and Dr E. K. Marshall for advice, encouragement and many helpful suggestions in regard to this work.

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THE UTILIZATION OF CARBOHYDRATES IN A CASE OF CHRONIC PENTOSURIA

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(Received for publication April 23, 1926)

Recently an opportunity was afforded for a brief study of the carbohydrate and respiratory metabolism in a case of chronic pentosuria. Simultaneous blood sugar and respiratory quotient time curves were obtained following ingestion of glucose as described hereafter. As far as can be ascertained from the literature, such observations, in this rare disease, have not been recorded hitherto.

The subject, a male, aged 40, applied recently for a life assurance policy, and was rejected because sugar was found in his urine. Otherwise he was perfectly well and there were no signs suggesting diabetes mellitus. The applicant failed to mention the fact to the medical examiner, that he had had sugar in his urine since 1904 (for 22 years), at which time it was discovered accidentally, also during a life assurance examination. He also failed to present a letter (which he did to the author), given him by his physician at that time, showing that a diagnosis of pentosuria had been made.

Though alimentary pentosuria is not infrequently met with, chronic (essential, spontaneous) pentosuria is rare. Less than fifty cases are on record. Though relatively much has been written upon this condition, what is actually known is briefly and very clearly described in the recent edition of Garrod's classical monograph on "Inborn errors of metabolism" (1). It is therefore unnecessary to discuss in detail the various aspects of our knowledge of this disease. Only the essential features of clinical interest will be mentioned briefly.

The condition belongs to the group of "inborn" diseases, and is characterized by the presence of pentoses in the urine which bear no relation to the ingestion of these sugars. Of the forty-four cases recorded by Garrod, thirty-three were males. The youngest was a

heating, and then becoming apparent suddenly. This delayed reduction of Fehling's solution, frequently mentioned in text-books as being of diagnostic value, is not necessarily due to pentose, and is therefore unreliable for this purpose. Garrod points out that urines containing small traces of glucose may behave in the same way, and quotes Bial's observations that it may be observed when the urines have been kept for some time with addition of preservatives, such as toluene or chloroform. The pentose usually found in chronic pentosuria is not fermentable by yeast and is optically inactive. Phenylhydrazine produces an osazone, with a melting point of 156° to 160°C , and a nitrogen content of 17.07 per cent. The special color reactions (red with phloroglucinol and green with orcin) are not obtained with glucose. Both of the colored substances are soluble in amyl alcohol, and the alcoholic solutions show characteristic spectroscopic bands. In the phloroglucinol test the bands lie between the Fraunhofer lines D and E (about $\lambda 5425$ to $\lambda 5750$) and in the orcin test between C and D ($\lambda 5850$ to $\lambda 6150$). Glycuronates give similar color reactions, but they may be eliminated from the urine by controlling the ingestion of substances yielding glycuronates. The compound glycuronates are optically active.

The physical signs in this case were negative and for brevity are omitted. A careful inquiry, with reference to the family history, failed to find any other member of the family similarly affected. The urine reduced Benedict's solution, did not ferment with yeast, and was optically inactive. Positive results were obtained both with the phloroglucinol, the old orcinol and Bial tests. The melting points of the osazone crystals obtained with phenylhydrazine on three different occasions were 158.5° , 159° , 158° , respectively. Quantitative determinations of the daily excretions were made by the sulphite-iodometric method described by Cammidge and Howard (2) and on three successive periods the results were as follows, 3.6, 2.5, 4.0 grams—an average of 3.3 grams per day. During the last period the diet was very high in carbohydrates. It will be noted this did not alter the amount of pentose excreted. Ten units of insulin had no effect upon the rate of excretion on the one day in which it was given. No excess excretion of urobilin was found in the urine by the method of Wallace and Diamond (4). The Van den Bergh reaction for bilirubin in the

child five years old (Aron) Garrod points out that Alexander's case, a child of eighteen months of age, was probably of the "alimentary" type Cammidge and Howard (2) record a case of a female, 75 years old It tends to occur in several members of a family, but neither the parents nor offspring of such individuals are likely to be similarly affected—in this way differing from diabetes mellitus and renal glycosuria The Hebrew race appears to be particularly liable The condition is apparently harmless Garrod mentions two individuals (Klercker) observed for ten years Cammidge and Howard (2) record one who was treated for twenty-six years for diabetes One subject reported by the late Theodore C Janeway (3) in 1906 was recently met with by the writer¹ Having applied recently for life insurance, the company insisted upon a "sugar tolerance test," the results of which, however, indicate that his glucose metabolism is not perfectly normal In the post absorptive state the blood sugar was 0.085 per cent Following ingestion of one hundred grams of glucose, the blood sugar rose to 0.241 per cent at the end of thirty minutes, but returned to the original level in two and a half hours In the case which is the subject of this report, the diagnosis, as just stated, was made twenty-two years ago (1904)

The amount of sugar excreted is usually small—about 5 grams—and is not affected by the ingestion of glucose yielding carbohydrates Low protein and purin free diets have been noted to lessen excretion Pentoses or pentose yielding foods ingested in the usual quantities, appear to be destroyed the same as in normal individuals In the one case mentioned by Garrod (Blumenthal and Bial) in which an examination was made of the blood sugar, the latter was found to be normal Cammidge and Howard (2) found excess urinary excretions of urobilin and amino acids in all of their seven cases, and therefore suggested defective liver function as an underlying cause

DIAGNOSIS

Because arabinose does not reduce Fehling's solution as readily as glucose, and the amount of sugar is usually small, the reduction of this solution may be delayed—taking place some time after the second

¹ I am indebted to Dr C C Birchard, Chief Medical Officer of the Sun Life Assurance Company of Canada, for having brought this case to my attention

study of renal glycosuria (5), (6), and therefore need not be described here. The combined results are shown in table 1.

DISCUSSION OF RESULTS

The blood sugar in the basal state was normal (0.080 per cent). Following the ingestion of 100 grams of glucose the maximum concentration (0.135 per cent) was noted at the end of one hour, and the original concentration was again reached before the end of the third hour. From these observations there was no indication of diminished sugar tolerance.

The total respiratory quotient in the basal state was 0.822. A fall was noted at the end of the first hour. This phenomenon was previously (6) discussed and is compatible with the normal. The maximum increase (0.054) was noted at the end of the third hour. The maximum percentage increments above the basal, of carbon dioxide production, oxygen consumption and heat production were 13.2, 9.6 and 10.2 respectively. The total amount of glucose oxidized during the three hour period was 21.99 grams or an average of 7.33 grams per hour.

All these values correspond to the normal. Confirmation, by modern methods, is therefore given to the general opinion held that in chronic pentosuria (unlike diabetes mellitus) the utilization of carbohydrates is unimpaired. There appears, therefore, to be no reason for either subjecting these individuals to restricted diets or classifying them as sub-standard insurance risks.

Grateful acknowledgment is due Miss Althea B. Frith and Miss Eleanor V. Bazin, who were responsible for all blood and respiratory air analyses.

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blood plasma was negative (direct and indirect) There was therefore no evidence in this case, as in those of Cammidge and Howard, of defective liver function

In the post absorptive state, on February 18, 1926, the blood sugar was normal (0.078 per cent) One hundred grams of glucose were then given by mouth with the following blood sugar results

	<i>per cent</i>
At the end of 30 minutes	0.089
At the end of 60 minutes	0.092
At the end of 120 minutes	0.086
At the end of 180 minutes	0.066

TABLE 1
Simultaneous respiratory exchange and blood sugar time curve data

Time	Liters		Nitrogen per hour (grams)	R Q		Total calories per hour	Grams of carbohydrate oxidized per hour	Blood sugar, per cent	Percentage increase above basal		
	O ₂ per hour	CO ₂ per hour		Total	Non protein				Oxygen consumption	CO ₂ elimina- tion	Heat produc- tion
Basal	13.68	11.24	0.402	0.822	0.824	66.1	5.93	0.080			
30 minutes								0.104			
60 minutes	14.82	11.57	0.430	0.781	0.776	70.8	4.04	0.135	8.3	2.9	7.1
120 minutes	15.00	12.73	0.444	0.849	0.858	72.9	8.66	0.113	9.6	13.2	10.2
180 minutes	13.38	11.72	0.400	0.876	0.891	65.6	9.29	0.068		4.2	

Sugar was found in the urines obtained at the end of each of the above periods. All specimens, with the possible exception of one, were, however, optically inactive, were not fermented by yeast, and gave a positive Bial reaction. In the one exception a small quantity of gas (equal to that from 0.25 per cent glucose) was found in the saccharometer, which did become smaller on the addition of sodium hydrate.

These observations alone suggested no disturbance in the ability of this individual to metabolize the ordinary carbohydrates. His dietary habits are such that he takes the normal quantity of carbohydrates per day, and has no particular tendency to take pentose yielding fruits and vegetables. He returned for further observation on April 13, 1926, and at this time blood sugar and respiratory exchange time curves were obtained simultaneously. The procedures followed were the same as employed previously in this laboratory in a similar

A NEW TYPE OF OXYGEN CHAMBER

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(Received for publication April 26, 1926)

During the last decade the therapeutic use of oxygen has received considerable attention. The physiologic basis for its use has been reviewed by Means (1) and by Lundsgaard (2). In a recent communication (3) we have discussed the efficacy of various types of portable apparatus and the clinical results obtained from adequate oxygen therapy. We wish now to report a new type of oxygen chamber in which the ventilation is secured by a thermal circulation of the air.

An oxygen chamber may be defined as a room which is leak-tight in order to provide an atmosphere rich in oxygen and which is artificially ventilated in order to maintain a comfortable environment. The oxygen content of the room is raised until the concentration of oxygen is between 40 and 60 per cent, which is considered the therapeutic range. It is ventilated in order to remove excess carbon dioxide, moisture and heat. Provision is made to keep the CO_2 concentration below 1.0 per cent, maintain a temperature between 58° and 68°F , and secure a relative humidity between 50 and 70 per cent.

These conditions have been met previously by the construction of a room within a room. The inside room, or oxygen chamber, is ventilated by means of motors, one to four in number, which operate a corresponding number of air pumps or fans. In addition, a refrigerating plant or frigid air system is present for removal of heat and moisture. All the machinery is kept outside the chamber, at times in a separate room. The air is pumped out of the chamber, passed through soda-lime and over coils of ammonia or brine, and then returned to it (4).

The chamber which we are reporting differs from the above in that there are no pumps, fans, motors, artificial refrigerating plant, or any electrical appliance. In addition, the chamber is transportable. It is

lined with a rubber gasket and the adjoining panels bolted together. This method of construction applies through the chamber with the exception of the floor and doors which are reinforced by strip aluminum rods.

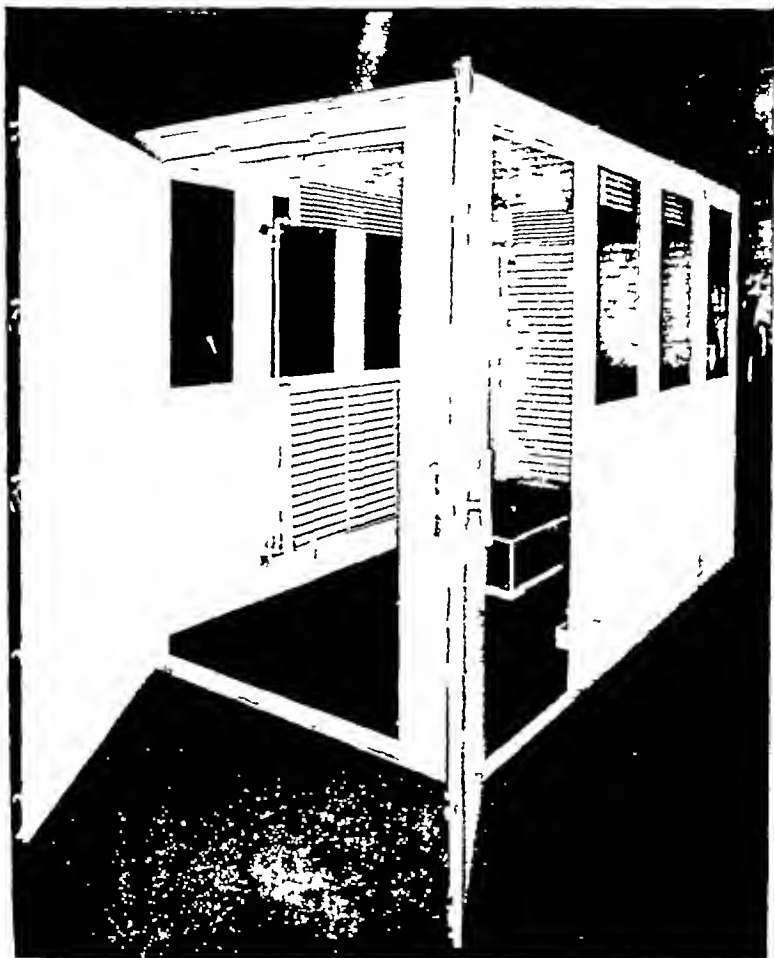


FIG. 1 OXYGEN CHAMBER, SHOWING TWO DOORS, COILS AND SODA-LIME BOX

The inside of the chamber contains several banks of coils, the disposition of which is seen in figures 1 and 2. The head end contains

constructed of aluminum and glass panels, which are joined together with the intervention of rubber gaskets and bolted in place. The inside of the chamber is lined in certain places by aluminum pipe which contains circulating cold water from the ordinary cold water faucet. The water enters the pipe at one side of the chamber and leaves at the other. The exit water returns to the drain. A large soda-lime container is placed under the bed. No other apparatus is used.

The air in contact with the cold aluminum pipes is chilled, the moisture is condensed on the surface of the pipes, and the cool dry air passes to the floor of the chamber where the carbon dioxide is removed by contact with the soda-lime. The air is warmed by the heat of the patient's body and passes to the roof with additional moisture and carbon dioxide. It is then chilled again and the process is repeated. By the use of these convection currents there is obtained an adequate removal of carbon dioxide, moisture and heat.

CONSTRUCTION OF THE CHAMBER

The dimensions of the chamber are 10 feet long, 7 feet high and $5\frac{1}{2}$ feet wide. The two side walls are composed of panels 7 feet high and $2\frac{1}{2}$ feet in width. The head end contains two panels 7 feet high and $2\frac{3}{4}$ feet wide. The foot end is made in a solid piece to enclose a door $6\frac{1}{2}$ feet high and 44 inches in width. The floor and roof of the chamber are composed of panels $5\frac{1}{2}$ feet long and $2\frac{1}{2}$ feet wide. In one of the side panels nearest the foot end a smaller door has been constructed $6\frac{1}{2}$ feet high and 2 feet wide. The remaining side panels have a window in the upper half, 3 feet high and 2 feet wide. The glass is of the triplex variety, i e, two thicknesses of glass with celluloid between, which is exceedingly strong and does not splinter.

The aluminum panels have to be made with especial care to obtain strength, lightness, durability and a leak-tight juncture. An aluminum sheet of the dimensions of the panel desired is welded on all four sides to an aluminum angle. The sheet is 0.030 of an inch in thickness, whereas the angle is 0.25 of an inch in thickness. The welding of materials of such difference in thickness is very difficult and requires a highly trained aluminum welder. The angle is 1 inch by 1 inch. Each aluminum sheet is thus set in a frame which consists of the aluminum angle. The side of the angle which projects outward is then

adjacent to the foot end are free from any coils. The pipe is $\frac{1}{2}$ inch in outside diameter. Each bank of coils is equipped with a valve so that they may be used all together or one at a time as is desired. At the bottom of the coils is an aluminum trough which conveys the condensed water to the outside by means of a pipe water-sealed.

The soda-lime box is 4 feet long, 2 feet wide and 1 foot high and is made of copper screening, supported by aluminum rods 1 inch in width. Two trays of the same material are suspended in the box to increase the area of soda-lime exposed. The box is placed at the head end of the chamber underneath the bed.

Four $\frac{1}{4}$ -inch holes are placed at the head end of the chamber for withdrawal of air for testing of oxygen and carbon dioxide. The simplified oxygen analyzer described by B nger (4) is used and an early Henderson modification of Haldane's CO₂ analyzer. For routine use Marriott and Howland's phenolsulphonephthalein test is employed for determining CO₂ concentration. Two $\frac{3}{8}$ -inch holes are placed in the head end for admission of oxygen.

THE OPERATION OF THE CHAMBER

The assemblage of the chamber is accomplished simply by bolting the panels in place in their respective positions. When a patient is put in the chamber the large door is used, and either the patient is transferred on a stretcher to a bed in the chamber or the bed containing the patient is rolled in. A can of soda-lime is emptied in the box, 50 pounds being distributed in three layers. This is sufficient to last approximately one week. If it is planned to keep the patient in the chamber two weeks two cans may be emptied in the soda-lime box. The valve regulating the inlet of cold water is turned on full, and may later be regulated to a slower flow if the room temperature falls below 58°F. The oxygen is first admitted at the rate of 30 liters per minute until the concentration reaches the desired level, and then adjusted to between 6 and 10 liters per minute depending on the amount required.

The small door at the side is used for entrance of doctor and nurse. A curtain of rubberized silk is suspended inside this door to prevent loss of oxygen when entering or leaving the chamber. When leaving the chamber it is advisable to get between the curtain and the door

a solid bank of pipe from the roof to 8 inches from the bottom and extending in width to within 6 inches from each wall At the sides

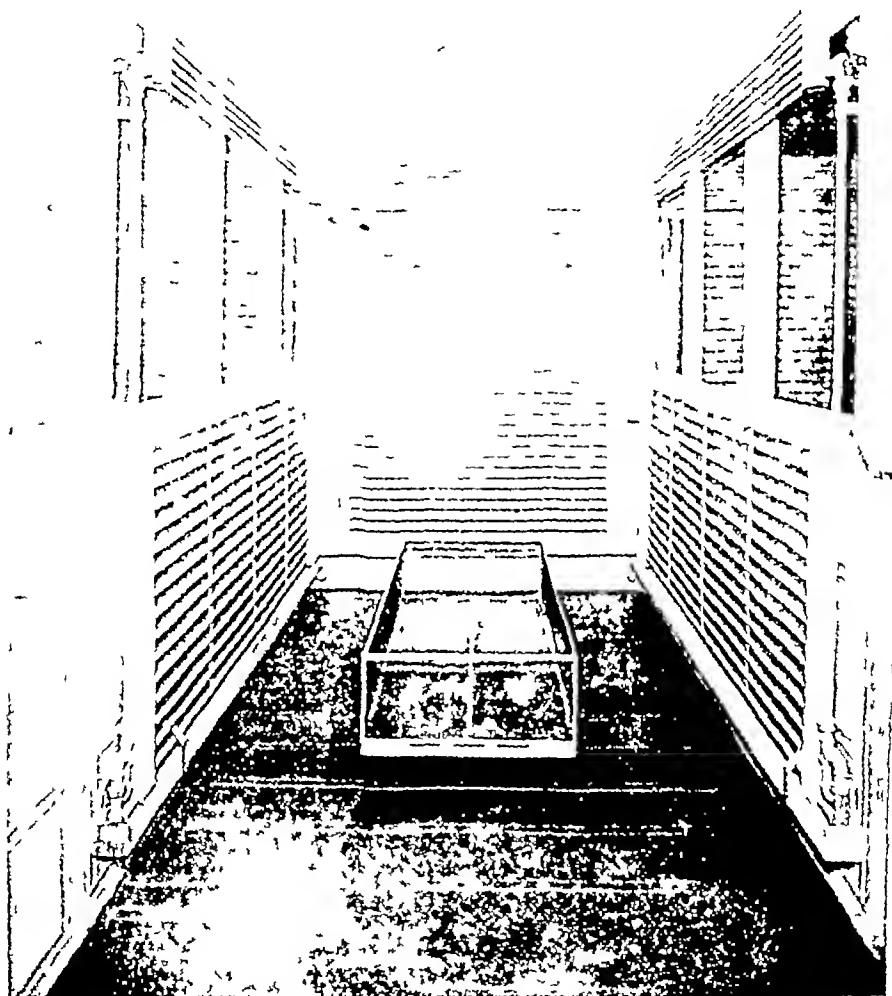


FIG 2 INTERIOR OF OXYGEN CHAMBER

Pipes line three walls Soda-lime box is placed under the bed

there are two banks of coils, one at the top, $1\frac{1}{2}$ feet in height and 7 feet in length, and one at the bottom, 3 feet in height and 7 feet in length The foot end of the chamber and both sides for a distance of 3 feet

before opening the door, allowing the curtain to block the free diffusion of air and oxygen when the door is opened

The inflow of oxygen is regulated by a specially calibrated reducing valve which is connected by a manifold to five high pressure oxygen tanks. Each tank contains 6000 liters. The valve is calibrated in liters per minute. The amount of oxygen needed for a given length of time can be calculated and the oxygen tanks renewed accordingly.

VENTILATION DATA OF THE CHAMBER

Good ventilation depends upon the maintenance of an atmosphere of such temperature, humidity and state of motion as will cool the human body without chilling it. The standard in use under ordinary circumstances includes a temperature of from 65° to 70°F, a humidity between 50 and 70 per cent, and an absence of drafts of cold air. The oxygen and carbon dioxide concentrations are no longer considered criteria of the hygienic purity of the air as these factors do not play a rôle until long after the atmosphere has become oppressive from other causes, such as heat, moisture and odor.

Human comfort is dependent upon the rate of heat production within the body and the cooling power of the air. There is a constant loss of heat from the surface of the body by radiation, convection and evaporation. Lowering the temperature of the air increases the cooling power by accelerating radiation and convection, whereas lowering the humidity increases the cooling power by accelerating evaporation. The health, comfort, and physiological efficiency of men exposed to atmospheres of varying temperature and humidity have been investigated by the U. S. Bureau of Mines in cooperation with the American Society of Heating and Ventilating Engineers (5). They have found that certain combinations of temperatures and humidities will produce the same total body heat loss, by radiation, convection and evaporation, and therefore the same feeling of comfort and discomfort. Lines passing through such air conditions, plotted in the form of a psychrometric chart, they have designated equal comfort or effective temperature lines (6). Thus, a temperature of 65°F with a relative humidity of 100 per cent is equally as comfortable as a temperature of 70°F with 50 per cent relative humidity or a temperature of 78°F with 0

relative humidity. This particular line they have designated the comfort line for still air conditions.

Effective temperature appears to be a very valuable term for defining conditions of the atmosphere inasmuch as it represents the cooling power of the air and is capable of exact definition. For patients with fever we believe the effective temperature should be lower. (See table 1 for ventilation data.) The temperature of the chamber is kept at 60°F with a relative humidity of 60 per cent, which corresponds on the effective temperature line to a temperature of 57°F with 100 per cent humidity or a temperature of 66°F with zero humidity. By regulating the flow of cold water and the number of banks of coils employed both the temperature and humidity may be lowered or increased.

Leonard Hill (7) has made an exhaustive study of the science of ventilation through the development of a new instrument, the kata-thermometer. The cooling-power of the air and the velocity of air movement may be determined by its use. As these factors are of profound importance in estimating good ventilatory conditions the kata-thermometer was used to determine the effectiveness of the chamber in these respects.

The kata-thermometer is a specially constructed alcohol thermometer with a cylindrical bulb 1.8 cm. in diameter and 2.2 cm. in length. The stem, 20 cm. in length, is graduated in tenths of a degree Fahrenheit from 100 to 95. The procedure followed in taking a reading is to heat the kata in water until the alcohol rises to the top. The bulb is then dried and the instrument set up firmly in a suitable position. The time is taken for the fluid to fall from 100 to 95, measured by a stop-watch. This time is a function of the heat loss from the surface of the kata by radiation and convection. To take the wet readings a muslin finger-stall is put on the bulb, and the same procedure followed. The wet kata gives the heat loss from radiation, convection and evaporation. As the total heat lost from the surface of the kata in cooling from 100 to 95 is always the same, the rate of heat loss depends entirely upon the surrounding atmospheric conditions, namely, temperature, humidity, and air movement.

The total heat loss in cooling from 100 to 95, determined experimentally in millicalories, divided by the surface area of the kata gives

before opening the door, allowing the curtain to block the free diffusion of air and oxygen when the door is opened

The inflow of oxygen is regulated by a specially calibrated reducing valve which is connected by a manifold to five high pressure oxygen tanks. Each tank contains 6000 liters. The valve is calibrated in liters per minute. The amount of oxygen needed for a given length of time can be calculated and the oxygen tanks renewed accordingly.

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a good standard of comfort for office and residential buildings. This would ordinarily require an open window or some system of artificial ventilation.

The efficiency of the chamber was determined from this point of view and the data tabulated in table 1. It is seen that the cooling power varied between 5.8 and 7.3 millicalories per square centimeter per second, with an average approximately of 6.5. The cooling power is about 1 millicalorie higher at the floor than at the roof. This is explained by the temperature being 7 degrees lower at the floor than at the roof. The relative humidity is about 10 per cent greater at the floor due to the lower temperature.

TABLE 1
Air velocity, cooling power, temperature and humidity of oxygen chamber

Measurement of	Head-end wall		Foot-end wall		Right wall		Left wall	
	1 foot from roof	1 foot from floor	1 foot from roof	1 foot from floor	1 foot from roof	1 foot from floor	1 foot from roof	1 foot from floor
Temperature of dry bulb	63.0	55.8	63.0	56.2	64.0	56.5	62.5	56.3
Temperature of wet bulb	54.3	49.8	54.0	50.0	54.0	51.0	54.5	50.0
Relative humidity	56	65	55	64	51	68	59	65
Dry kata T	76	62	74	63	78	63	72	62
Wet kata T'	36	33	38	34	39	34	39	34
Cooling pH	6.0	7.3	6.1	7.2	5.8	7.2	6.3	7.3
H	0.173	0.175	0.177	0.174	0.173	0.175	0.180	0.177
$\Phi - t$								
Velocity, feet per minute	14.5	15.5	16.5	15.5	14.5	15.5	18.0	16.5

Note. In each instance measurements were taken 4 inches from the wall at its center.

The heat loss from the wet kata is 12.6 millicalories per square centimeter per second. The cooling effect according to Hill could be roughly analyzed as following: 6 millicalories by evaporation, 3.3 millicalories by radiation and 3.3 millicalories by convection.

In a room which we have tested the dry kata heat loss was 3 millicalories with the window closed. The air velocity was too low to be measured. With the window open the cooling power increased to 6 and with the window widely open much higher figures were obtained, with parallel increase in wind velocity.

The velocity of air movement in the chamber varied from 14.5 to

the heat loss in millicalories per square centimeter. This figure is known as the "kata factor," and constitutes the converting constant of the reading of any kata-thermometer in millicalories per square centimeter.

Thus, if F equal the total heat loss per square centimeter and T equal the number of seconds taken by the kata to cool from 100° to 95°F in an atmosphere with a temperature t , the heat loss can be expressed according to the law of cooling of hot bodies by the formula

$$F = K (\phi - t) T \text{ or in an individual instance, } \phi \text{ being } 36.5, \\ F = 0.27 (36.5 - 17.8) 105.9 = 535, \text{ or the kata factor}$$

The rate of cooling per square centimeter per second is then given by

$$H = \frac{F}{T} \text{ or } K (\phi - t), \text{ in millicalories}$$

To calculate the wind velocity from the dry kata reading the following formula is used

$$\frac{H}{(\phi - t)} = 0.27 + 0.49 \sqrt{V}$$

V can be determined in meters per second or feet per minute. The above explanation indicates the method used and will suffice for the purposes of this paper. A more detailed study may be found in Hill's book or in a recent paper by McConnell and Yaglaglou (8).

Air movements have been explored by means of mechanical vane anemometers and by the inclination from the vertical of paper flags or silk threads, or by the drift of small balloons. As Hill says, none of these methods appreciates as the kata does every eddy, no matter how small and brief, which go to make up the sum total of the movements which affect the skin. "Thus fallacious results may be obtained on comparing the air movement in a room ventilated by the plenum system, which is more unidirectional in character and affects flags, and the movement in a room with open windows in which there are eddies, which neutralize any steady inclination of flags. The kata, then, seems to be a suitable instrument for measuring the physiological efficiency of air movement in rooms." He believes that a dry kata heat loss of 6 millicalories per square centimeter per second constitutes

the general movement of the air in the manner cited may be seen by blowing cigarette smoke against the various walls (This experiment, it need hardly be cautioned, may be performed only when no oxygen has been added to the air)

The carbon dioxide is removed as the air comes in contact with the soda-lime under the bed. The soda-lime box is made of copper screening of fine mesh, strengthened by strips of aluminum 1 inch in width. Two additional trays have been added to increase the speed and area of absorption although they are not really necessary. Fifty pounds of soda-lime last approximately one week. It is perfectly feasible to place enough soda-lime in the box to last three weeks of continuous use without stirring or renewing. The CO_2 concentration is maintained below 0.5 per cent, although 1 per cent may be allowed as a maximum. Wilson soda-lime, 4 to 8 mesh, 15 per cent moisture should be employed. This is a very efficient carbon dioxide absorber and is sufficiently porous to allow the air to pass through it as well as over it. Fine mesh soda-lime, however, prevents free passage of air and is thus unsatisfactory for this purpose.

The condensation of water on the pipes has an additional advantage beyond that of maintaining the humidity within normal limits. Odors are apt to be condensed with the water, so that the chamber is absolutely free from all skin odors, as well as those which occur from excreta.

The temperature of the chamber may readily be kept 10 degrees lower than the atmosphere surrounding it. Thus, the room enclosing it in the hospital is maintained at 70°F and the chamber at 60°F. During the summer the water from the faucet may be 8° warmer than it is during winter, so that a temperature of 68° to 70°F may obtain in the chamber. If more cooling is desired the water before entering the chamber will be made to pass through a copper coil packed in ice. No difficulty is anticipated although it has not as yet been used in the summer months. As a matter of fact, cooling in this type of chamber is as easily secured as those employing a unidirectional system of ventilation in which the air has to be taken out of the chamber and cooled.

The chamber is leak-tight when in use. In chambers in which the

18 0 feet per minute The margin of error is such as to include these variations, so that it may be said that the air velocity is approximately 16 feet per minute The type of air movement corresponds to the window-ventilated room where the cold air comes in at the bottom and rises to the ceiling as it is warmed, differing radically from a unidirectional system of ventilation In figure 3 a diagrammatic sketch is given of the air movement when the coils at the head-end and at

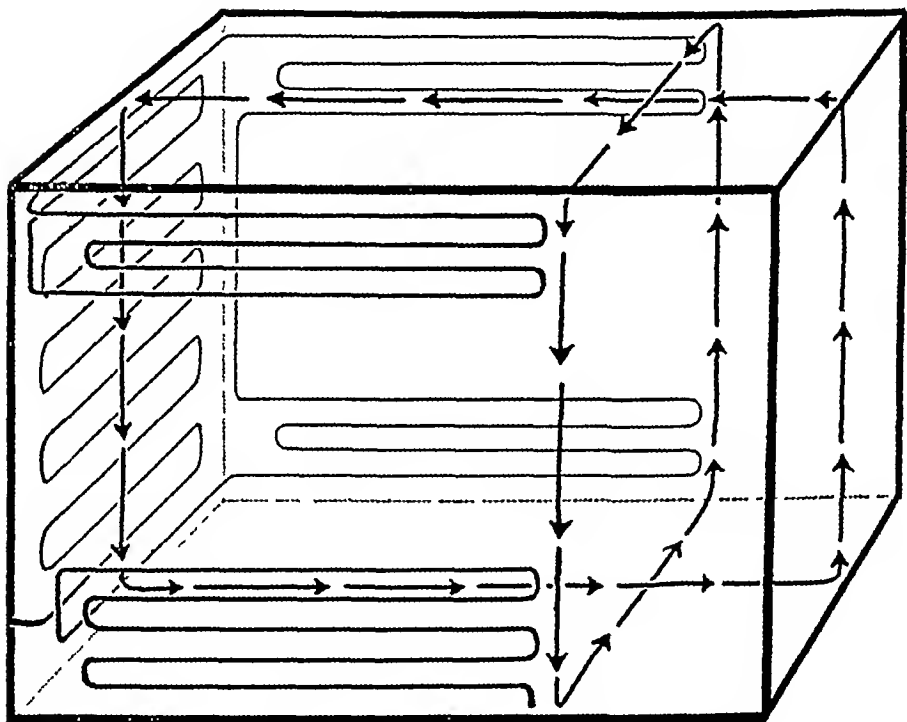


FIG 3 DIRECTION OF TWO MAIN CONVECTION CURRENTS IN OXYGEN CHAMBER INDICATED BY ARROWS

one side are in use The air at the head-end moves downward, traverses the floor underneath the bed, ascends the opposite end and returns along the roof to the head-end Another current of air passes downward from the coils on one side to the floor, passes across to the opposite side, ascends to the roof and returns to its starting point The heat of the patient's body is responsible for the warming of the air Numerous eddies and cross-currents are naturally present, but

namely, at 57°F The dry kata heat loss or cooling power is 6 mill-calories per square centimeter per second The velocity of air movement is 16 feet per minute

4 The chamber is transportable It is constructed of aluminum and glass panels in such a way as to be easily demounted and assembled without destroying its leak-tight property

5 The operation of the chamber is simple and economical. By turning on the water and the oxygen the running of the chamber is initiated and maintained The expense of up-keep is six to eight dollars a day¹

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¹ This oxygen chamber may be obtained from the Paramount Welded Aluminum Products Co., 798 Wythe Avenue, Brooklyn, N Y

air has to be removed by pumps or fans leaks are apt to develop in the connections, the soda-lime containers or the refrigerating apparatus, or in the pumps themselves. With care these leaks can be removed or prevented, but our experience with the ventilation of tents has taught us that constant care is necessary. In this chamber in which all ventilation is secured inside the chamber leaks from this source do not occur.

The expense of the chamber is thus limited to the cost of the oxygen and soda-lime, which amounts to six to eight dollars a day. High pressure oxygen is employed (Linde Air Products). A reducing valve is used, specially calibrated to deliver oxygen in liters per minute (Oxweld Acetylene). Five 220 cubic foot tanks are connected to the valve by means of an oxygen manifold.

The chamber has been used in fifteen cases up to the present. Our intention is not to report the clinical results at this time but merely to state that it has proved satisfactory from the various points of view mentioned above. The management of the patient both from a nursing and medical point of view is unhampered. There is no machinery to get out of order. There is no source for sparks as no motors or pumps are employed. The running of the chamber is initiated and maintained by turning on the water and the oxygen.

SUMMARY

1 A new type of oxygen chamber is described in which there is adequate removal of carbon dioxide, moisture and heat without the use of motors, fans, electrical appliances or artificial refrigerating systems.

2 The inside of the chamber is lined in certain places by aluminum pipe which contains circulating cold water from the cold water faucet. The air in contact with the cold aluminum pipes is chilled, the moisture is condensed on the surface of the pipes, and the cool dry air passes to the floor of the chamber where the carbon dioxide is removed by contact with soda-lime. The air is warmed by the heat given off from the body of the patient and the process is repeated.

3 Data are presented which demonstrate that the chamber fulfils the modern requirements of scientific ventilation. Its effective temperature can be maintained below that ordinarily deemed necessary,

STUDIES IN BLOOD VOLUME

I THE BLOOD VOLUME IN MYXEDEMA, WITH A COMPARISON OF PLASMA VOLUME CHANGES IN MYXEDEMA AND CARDIAC EDEMA^{1 2}

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(Received for publication April 28, 1926)

I INTRODUCTION AND LITERATURE

One of the first things that patients with myxedema notice when thyroid extract is administered to them, is an increase in water intake and urinary output. The diuretic action of thyroid substance was indeed, one of the first effects of its use observed, and was demonstrated as early as 1890 by Leichtenstern (1). Since then several observations have been made which support the idea that the thyroid gland is a factor in maintaining the water balance.

Gardella (2) in 1910, and Paladino (3) in 1912, both found increased viscosity and decreased electrical conductivity of the blood serum in dogs and rabbits following thyroparathyroidectomy. A few years before this, however, Fano and Rossi (4) demonstrated that the viscosity of serum, while increased from thyroparathyroidectomy in animals, was not increased after extirpation of the parathyroids alone. Kottmann (5) and Deusch (6), several years later, and Neuschlosz (7) more recently, reported that increased viscosity of the serum occurred in patients with myxedema. From refractometric measurements, Deusch further concluded that the total protein of the serum is

¹ This paper was read at the meeting of the Society for Clinical Investigation in Atlantic City, May 3, 1926.

² This study was aided by a grant from the Proctor Fund of the Harvard Medical School for the Study of Chronic Diseases.

³ Research Fellow in Medicine, Harvard Medical School and Massachusetts General Hospital.

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An observation of some interest is that of Wilhelmj and Fleischer (14), (15) that thyroxin fed to normal guinea pigs causes an average decrease in surface tension of plasma of 3.7 dynes and that thyroidectomy in these animals causes an average increase in surface tension of plasma of 5.1 to 5.7 dynes. Thyroxin added directly to plasma had no effect.

Two years ago Hildebrandt (16) made the very important observation that the intravenous injection of 1 mg. of thyroxin in rabbits after a hunger-thirst period of 14 to 15 hours caused marked hydremia with a maximum in the fifth or sixth hour, at which time the total quantity of blood was usually raised 40 per cent above the normal (as measured by hemoglobin and red blood cell dilution). Parallel to this occurrence there was a large water and salt diuresis. The normal red count was reached again after 36 hours.

In support of the idea therefore, that the thyroid gland is concerned in water balance, there are the following observations:

- 1 Thyroidectomy in various animals causes
 - a Increased viscosity of the serum
 - b Decreased electrical conductivity of the serum.
 - c Increased refractive index of the serum
 - d Decreased water per cent of serum.
- 2 Thyroid insufficiency in human beings causes
 - a Increased viscosity of the serum
 - b Increased refractive index of the serum.
- 3 Intravenous injection of thyroxin in normal rabbits produces
 - a Marked hydremia
 - b Pronounced water and salt diuresis

Thus far no direct observations of the total quantity of circulating blood or plasma have been made in disorders of the thyroid gland. Since changes in viscosity, refractive index, electrical conductivity and water percentage of serum, and changes in hemoglobin concentration and red blood cell count may occur without changes in the total quantity of serum or plasma it is important to learn whether there is a relation between them and disease of the thyroid gland. The fact that in experiments thus far reported the results all point in the same direction certainly suggests and points to the fact that the serum and plasma are concentrated in myxedema.

increased from 5.3 to 9.1 per cent. Hammett (8) has suggested that this interpretation should be accepted with reservations in view of the finding of Peterson, Jobling and Eggstein (9) of a progressive increase in the non-protein nitrogen and proteoses after thyroidectomy in animals and of Étienne, Richard and Roesch (10) of an increase of urea nitrogen in the blood in thyroid insufficiency. The increase in all three factors is, however, more marked in experimental myxedema and in cretinism than in human myxedema and Deusch's conclusion that there is high serum protein in myxedema is probably correct.

Deusch also found the sodium chloride in serum to be within normal limits in myxedema and observed little change in this respect before and after thyroid administration. From this he concluded that salt and water excretion ran parallel with their removal from the tissues and that the water content of the serum was therefore unchanged by thyroid. He concluded that the increase in protein was an absolute and not a relative one.

In 1917 Eppinger (11) made an important contribution to the subject when he demonstrated that salt solution injected subcutaneously and water given by mouth are absorbed more slowly and excreted more slowly in thyroidectomized than in normal dogs, and more slowly in normal than in thyroid-fed ones. He also observed that in some cases of cardiac and nephritic edema when all known methods of causing fluid elimination had failed, the administration of thyroid substance produced marked diuresis. More recently Danzer (12) has reported that giving thyroid substance caused a distinct tendency to rapid absorption in three cases of pleural effusion following lobar pneumonia. The difficulty with such observations is that controls cannot be obtained. Loeb (13) however substantiates this view in a reference to the fact that Hoover found a collection of a certain amount of fluid in the peritoneal cavity in guinea pigs suffering from marked inanition, while it was absent in the large majority of animals in which the same or even a greater amount of inanition was produced through the combined effects of thyroid feeding and diminished intake of food.

Recently Hammett (8) has shown that loss of the thyroid gland in male rats 100 days of age, causes a decrease in the water percentage and an increase in the refractive index of blood serum.

cent from the average (table 1) All of the calculated total blood volumes were within 7 per cent of the average Blood for these observations was collected in the morning after a hunger thirst period of 14 to 18 hours, but was not preceded by a rest period The results serve to illustrate that total plasma and calculated total circulating

TABLE 1

The constancy of plasma and calculated total blood volumes in a normal man

Age 42 years, height 166 cm.

Date	Weight	R.B.C.*	Hb	Hematocrit	Plasma volume		Total blood volume	
					Amount	Per kilogram	Amount	Per kilogram
	kg	millions	per cent	per cent cells	cc	cc.	cc.	cc
1/15/24	61.4	4.8	98	54.2	2,585	42.2	5,650	92.2
1/21/24	60.4	4.5	99	58.6	2,080	34.5	5,030	83.5
1/24/24	61.4	4.9	92	49.2	2,555	41.6	5,030	82.0
1/29/24	62.7	4.3	85	47.2	2,775	44.3	5,270	84.0
2/ 2/24	63.4		94	50.0	2,800	44.2	5,600	88.5
2/ 7/24	61.1	4.7	91	49.9	2,520	41.2	5,030	82.3
2/15/24	60.7	4.6	100	49.4	2,635	43.3	5,200	85.6
(12 15 p.m.)								
2/15/24	60.7	4.8	98	50.0	2,480	41.0	4,970	82.0
(1 30 p.m.)								
2/19/24	60.4	4.1	95	48.3	2,865	47.4	5,550	92.0
(8 40 a.m.)								
2/27/24	60.9	4.0	99	52.0	2,730	44.9	5,690	93.5
(10 15 a.m.)								
2/27/24	60.9			51.5	2,755	45.2	5,680	93.3
(1 30 p.m.)								
3/23/24	58.9	4.6	94	47.9	2,510	42.6	4,810	81.8
4/ 6/24	59.8	4.6	93	48.6	2,635	44.0	5,125	85.7

* In this and the subsequent tables R.B.C. means red blood cells, Hb, hemoglobin, B.M.R., basal metabolism rate.

blood seem to be fairly constant in the same individual under approximately the same conditions Several similar results could be cited

In Lamson's (20), (21), opinion the vital red method is accurate for measuring plasma volumes, but he states that total blood volumes calculated from hematocrit estimations are not true measures of the total quantity of circulating blood because these differ when made on

In the fall of 1923, I began to make observations on the plasma volume in myxedema. This work was mentioned by Means (17) in his address before the Association for the Study of Internal Secretions in May, 1925. Nine patients have been studied, numerous observations having been made on each one (except patient 3) while myxedematous and while receiving thyroid. The cases have consistently shown the same results and a report now seems justified.

II METHOD

The plasma volume method of Keith, Rowntree and Geraghty (18) was used except that brilliant vital red, as suggested by Evans, was substituted for vital red and isotonic sodium oxalate as suggested by Hooper et al (19), was used instead of powdered oxalate, to prevent clotting. This method is generally recognized as being accurate within 5 per cent. In general it proved to be as accurate as this in my work. Total blood volumes were calculated from hematocrit values as determined from calibrated centrifuge tubes. Hemoglobins, where reported, were done by the Newcomer method. I found great difficulty in matching colors with this hemoglobin method and only the general trend of the values is of any significance.

In all of the patients the blood was collected in the morning and its collection was preceded by a hunger-thirst period of 14 to 18 hours. In all except the first three cases it was also preceded by a rest period of at least one-half hour. In all except the first two cases withdrawal of blood was preceded by a basal metabolism determination done with a Roth-Benedict apparatus.

Only typical cases of myxedema were selected for these observations and cases of so-called related conditions supposed by some to be of thyrogenous origin were omitted. Due to the scarcity of untreated patients with myxedema, it was necessary to use patients who had been previously treated.

III THE CONSTANCY OF THE PLASMA AND CALCULATED TOTAL BLOOD VOLUMES IN A NORMAL INDIVIDUAL

Of thirteen plasma volume observations made of a normal individual over a period of nearly three months, only two varied more than 5 per

TABLE 2
Summary of plasma volume and basal metabolic rate changes in nine cases of myxedema

Case number	Average off thyroïd				Average on thyroïd				Increase				Percentage increase			
	Plasma volume				Plasma volume				Plasma volume				Plasma volume			
	B M R	Per kilo-gram	Per square meter	cc	B M R	Per kilo-gram	Per square meter	cc	B M R	Total	Per kilo-gram	Per square meter	B M R	Total	Per kilo-gram	Per square meter
	<i>per cent normal</i>				<i>per cent normal</i>											
♀ 1		45.5	1,640	2,615		56.5	1,960	3,060		425	11.0	320		16.2	21.2	19.5
♂ 2		40.9	1,475	2,375		54.6	1,925	3,050		675	13.7	450		28.4	33.5	30.5
♂ 3	62	37.4	1,525	2,630	97	53.0	2,100	3,550	35	920	15.6	575	57	35.0	41.7	37.7
♀ 4	75	30.6	1,140	1,725	101	39.7	1,490	2,270	26	545	9.1	350	35	31.6	29.7	30.7
♀ 5	77	33.1	1,290	2,115	103	43.5	1,675	2,715	26	600	10.4	385	34	28.4	31.4	29.8
♀ 6	72	29.0	1,315	2,460	111	36.3	1,555	2,805	39	345	7.3	240	54	14.0	25.1	18.2
♀ 7	83	43.3	1,400	2,135	101	50.3	1,610	2,445	18	310	7.0	210	22	14.5	16.2	15.0
♀ 8	77	27.9	1,320	2,565	102	37.1	1,650	3,100	25	535	9.2	330	32	20.9	33.0	25.0
♀ 9	72	31.0	1,120	1,735	99	38.9	1,350	2,045	27	310	7.9	230	38	17.9	25.5	20.6
Average for group		35.4	1,360	2,260		45.5	1,700	2,780		520	10.1	340		22.9	28.5	25.2
Average for B M R cases	71	33.2	1,300	2,195	102	42.7	1,630	2,705	28	510	9.5	330	36	23.2	28.9	25.2

blood from different parts of the body and from the same part under different conditions. He states, however, that, under basal conditions, both plasma and calculated total blood volumes are fairly constant quantities and most of my observations in myxedema were made under basal conditions. Only 3 hematocrit estimations vary more than 3 per cent from the average of the group (table 1). This is a fairly typical example. The hematocrit readings of blood drawn before dye injection and that on blood withdrawn after dye injection usually varied less than 1 per cent from each other. No claim is made to accuracy for hematocrits as here determined. They appear, however, to be accurate enough to show gross changes in the volume of circulating cells (say 10 per cent and over) under basal conditions.

IV PLASMA VOLUME CHANGES IN MYXEDEMA

In all of the cases of myxedema, the administration of thyroid substance produced a well marked and permanent increase in the plasma volume, a decrease in the hematocrit reading and in the red blood count. In two of the cases the hematocrit value and red count subsequently increased due to an increase in the volume and number of circulating cells probably as a result of stimulating the bone marrow on administering thyroid extract. As a group, when not taking thyroid extract the average of the plasma volumes was low—35.4 cc per kilogram and 1360 cc per square meter, as compared with normal values of 42 to 52 cc per kilogram and 1500 to 2000 cc per square meter. The significant thing is, however, not the average change but that observed in each case because the myxedematous level in some was within the so-called normal limits. The change in these cases was, nevertheless, as great on the average as in the others. Attention is called to the fact that the increase in plasma was absolute and not merely relative due to loss of weight. Even cases 6 and 8 (tables 8 and 10), which lost weight when given thyroid extract showed total plasma increases of 14.0 and 20.9 per cent respectively. It is of interest to note that the change in total plasma volume in these two cases was less, however, than in the other patients who lost less weight. Both of these patients had slight pitting edema and marked dyspnea on exertion. The possibility of slight cardiac edema masking the myxedematous reduction in plasma volume is suggested.

1/15/24	58 1	1 61	60	-25	68	4 0	40 8	2,730	47 0	1,700	4,600	79 0	2,860
1/17/24	58 1	1 61	60	-25	70	4 2	46 2	2,165	37 7	1,355	4,030	70 1	2,520
1/21/24	57 5	1 60	45	-25	88	4 3	42 0	2,390	42 0	1,500	4,120	72 3	2,590
2/ 2/24	57 0	1 59	60		76	3 9	43 5	2,505	43 6	1,560	1,440	77 2	2,760
2/ 9/24	57 5	1 61	54		92	4 4	46 2	2,440	42 2	1,515	4,540	78 4	2,820
2/16/24	57 9	1 61	57	-21	84	4 3	43 2	2,485	42 3	1,535	4,385	74 7	2,710
2/20/24	58 6	1 62	57		95	5 1	50 9	1,920	33 1	1,195	3,910	67 5	2,430
3/22/24	58 0	1 61	63		89	4 0	38 2	2,440	41 6	1,505	3,950	67 5	2,410
4/ 2/24	58 6	1 62	55		80	4 1	38 2	2,540	42 2	1,560	4,110	68 3	2,525
4/14/24	60 2	1 63	49		82	3 2	39 9	2,520	43 1	1,555	4,190	71 7	2,580
4/18/24	58 4	1 62	55	-16									
4/25/24	57 1	1 59	74		70	3 2	32 8	2,940	51 4	1,850	4,370	76 5	3,030
5/ 2/24	55 9	1 58	82		71	3 4	35 3	3,095	55 4	1,955	4,760	85 6	3,020
5/ 6/21	57 5	1 60	80		55	3 5	38 5	3,150	54 8	1,970	5,120	89 2	3,200
5/12/21	57 5	1 60	80		75	3 6	38 3	3,140	55 3	1,975	5,090	89 6	3,200
5/20/24	56 8	1 59	68	+3	83		39 1	3,110	57 0	1,980	5,110	93 5	3,260
6/3 /21	54 5	1 57	88										
6/24/21	55 4	1 58	76		67	3 6	36 2	3,430	62 0	2,170	5,375	97 2	3,400
7/28/21	55 9	1 58	72		73	3 9	39 3	2,970	53 2	1,880	4,890	87 5	3,090
8/ 1/24	57 7	1 61	51		72	4 0	42 8	3,175	55 0	1,970	5,550	96 1	3,450
8/ 7/24	53 6	1 56	83		89	5 0	51 7	2,910	54 3	1,865	6,025	112	3,860
8/14/24	55 2	1 57	66		74	5 0	46 5	2,810	50 9	1,790	5,250	95 1	3,340
8/25/24	56 1	1 58	74		80	4 3	45 2	3,030	54 0	1,920	5,530	98 5	3,500
8/28/24	55 7	1 58	80		74	3 5	40 5	3,050	54 8	1,930	5,130	92 1	3,245
9/ 3/21	55 3	1 58	82		83	4 3	46 0	2,695	48 6	1,705	4,995	90 3	3,160
9/11/24	55 6	1 58	82		81	4 1	45 6	2,805	51 5	1,815	5,280	91 7	3,340

Began thyroid 6 grains daily April 16

Thyroid increased to 15 grains daily May 27
Decreased to 6 grains daily Increased to
15 grains daily June 17

Decreased to 5 grains daily July 7

Increased thyroid to 10 grains daily
August 20

Reduced thyroid to 5 grains daily

TABLE 4
Case of myxedema no 2
Male, age 37 years, height 161.5 cm

Date	Weight kg	Body surface sq m	Pulse	B M R per cent	HB per cent	R.B.C millions	Hematocrit per cent cells	Plasma volume			Total blood volume			Remarks
								Amount cc	Per kilogram cc	Per square meter cc	Amount cc	Per kilogram cc	Per square meter cc	
10/19/23					24	2.0								
10/22/23					46	2.3								
10/25/23						1.9								
10/27/23						3.3								
10/29/23						3.0								
10/30/23				-37										
11/13/23	58.2	1.61	70		60	3.7	30.6	3,140	54.0	1,950	4,530	77.9	2,820	November 5, started thyroid, 6 grains daily Increased to 15 grains daily today
11/18/23				-8										
11/23/23							29.5							
11/26/23				-18										Thyroid omitted November 24
11/27/23	55.4	1.58	80			3.5	34.7	3,205	58.0	2,030	4,910	88.9	3,110	
11/30/23	56.4	1.59	70				31.7	3,000	53.4	1,885	4,390	78.0	2,760	
12/3/23	57.8	1.61	75				33.5	2,280	39.5	1,420	3,430	59.5	2,130	
12/5/23	57.3	1.60	65				35.5	2,580	45.0	1,610	4,000	69.9	2,500	Started thyroid 5 grains t.i.d. December 6
12/8/23	56.3	1.59	70					3,330	59.2	2,095	5,200	92.4	3,270	Decreased to 6 grains daily December 12
12/17/23				-7	87	4.0	44.3	2,500	46.0	1,590	4,490	82.5	2,860	Often omits thyroid Thyroid omitted
1/7/24	54.5	1.57	80		79	4.5	42.1	2,065	37.8	1,315	3,570	65.1	2,275	
1/10/24	54.8	1.57	68	-5	70	4.3								

TABLE 6
Case of myxedema no. 1
1 emale, age 46 years, height 151 cm

Date	Weight kg	Body surface m ²	Pulse	B M R. per cent	R. B. C. millions	Hematocrit	Plasma volume			Total blood volume			Remarks
							Amount cc	Per kilogram cc	Per square meter cc	Amount cc	Per kilogram cc	Per square meter cc	
6/10/25	56.0	1.51	65	-18	4.1	39.1	12,270	40.5	1,505	3,730	66.6	2,470	On thyroid $1\frac{1}{2}$ grains daily Thyroid omitted
6/15/25	57.0	1.52	64	-28	4.5	42.5	1,865	32.8	1,225	3,240	56.9	2,130	
6/23/25	56.6	1.52	55	-25	4.8	47.0	1,735	30.6	1,403	2,75	57.8	2,155	
6/24/25	56.1	1.51	58	-26	4.6	49.0	1,680	29.9	1,110	3,295	58.7	2,180	Started on thyroid 6 grains daily
6/27/25	56.0	1.51	56	-24	4.6	51.0	1,715	30.6	1,353	3,800	62.5	2,320	
6/30/25	56.1	1.51	53	-25	5.1	45.6	1,775	31.5	1,703	2,640	57.9	2,150	
7/4/25	56.4	1.51	63	-19	4.4	43.7	1,890	33.6	1,250	3,360	59.7	2,225	Thyroid increased to 12 grains daily
7/8/25	57.4	1.52	71	-15	3.9	37.8	2,215	38.6	1,453	3,560	62.1	2,340	
7/11/25	57.1	1.52	71	-8	3.6	39.0	2,345	41.0	1,545	3,850	67.3	2,530	
7/18/25	56.7	1.52	75	-3	3.9	39.0	2,320	40.9	1,525	3,800	67.0	2,500	Thyroid decreased to $7\frac{1}{2}$ grains daily
7/25/25	56.5	1.52	80	+1	3.6	38.8	2,375	42.0	1,560	3,860	68.6	2,550	
8/1/25	57.5	1.53	80	-4		39.5	2,135	37.1	1,390	3,530	61.4	2,300	
8/8/25	56.9	1.52	74	-1	4.2	40.6	2,135	37.6	1,405	3,600	63.4	2,370	Thyroid increased to 12 grains daily August 10
8/15/25	57.3	1.52	82	+2	3.9	39.1	2,115	36.9	1,390	3,475	60.6	2,280	
8/20/25	57.5	1.53	83	-1	3.4	36.5	2,100	41.8	1,570	3,780	65.8	2,470	
9/10/25	57.3	1.52	82	+6	3.8	37.9	2,260	39.5	1,485	3,640	63.5	2,390	Ordered thyroid to be decreased to 12 grains again September 3 Patient not directions reversed Look thyroid 12 grains daily until September 3 and $7\frac{1}{2}$ grains daily since Increased to 12 grains daily Decreased thyroid to 6 grains daily Increased to $7\frac{1}{2}$ grains daily Increased to 9 grains daily
9/21/25	56.7	1.52	71	+6	3.8	36.9	2,330	41.0	1,530	3,690	65.0	2,430	
10/15/25	57.9	1.53	89	+7	3.9	37.9	2,235	38.6	1,460	3,595	62.1	2,350	
11/10/25	57.1	1.52	71	-1	4.3	39.4	2,340	41.0	1,540	3,860	67.6	2,540	
12/3/25	57.7	1.53	87	+3	4.0	39.1	2,270	39.4	1,480	3,745	65.0	2,450	

TABLE 5
Case of myxedema no 3
Male, age 62 years, height 159 cm

Date	Weight		Body surface		Pulse	B M R per cent	R B C per cent	HB millions	Hematocrit per cent cells	Plasma volume			Total blood volume			Remarks
	kg	lb	sq m	sq ft						Amount cc	Per kilogram cc	Per square meter cc	Amount cc	Per kilogram cc	Per square meter cc	
/ 1/24	70.9	155	1.74	18.8	60	38	57	95.7	44.1	2,570	36.3	1,475	4,600	65.0	2,640	Started on thyroid 15 grains daily Increased thyroid to 25 grains daily Decreased thyroid to 15 grains daily on November 16
/ 3/24	70.0	154	1.72	18.6	60		41	84.7	48.0	2,695	38.5	1,575	5,180	74.0	3,020	
/ 6/24	68.6	151	1.71	18.5	70	27	40	67.7	38.0	3,280	47.8	1,920	5,290	77.0	3,090	
/15/24	68.2	150	1.70	18.4	100	10	27	63.4	36.0	3,340	49.0	1,965	5,220	76.5	3,070	
/28/24	65.9	145	1.68	18.2	93	4	37	61.0	37.2	3,760	57.1	2,240	6,000	91.1	3,570	

10/ 2/25	62 9	1 63	70	0	3 6	30 2,600	41 41,595	3,725	59 3,2,290	Reduced to 4½ grains daily Increased to 6 grains daily Ordered to increase thyroïd to 12 grains daily December 15
11/ 2/25	63 4	1 63	80	+22	3 5	29 8,2,700	42 61,655	3,850	60 8,2,360	
11/20/25	62 7	1 63	65	-2	3 9	29 12,810	44 91,720	3,975	63 5,2,440	
12/ 9/25	61 7	1 62	66	+1	4 1	28 6,2,760	41 91,710	3,875	63 0,2,390	
12/24/25	61 3	1 61	76	+12	4 1	28 1,2,790	45 51,735	3,885	63 4,2,415	Thyroid reduced to 4½ grains daily
12/28/25	61 3	1 61	72	+14	3 8	28 3,2,820	46 01,750	3,935	64 2,2,410	
2/ 6/26	60 8	1 61	65	-2	4 1	29 2,2,625	43 21,630	3,715	61 1,2,300	

TABLE 7
Case of myxedema no 5

Female, age 42 years, height 157 cm

Date	Weight kg	Body surface sq m	Pulse	B M R. per cent	R B C. mil- lions	Hematocrit	Plasma volume			Total blood volume			Remarks
							Amount cc	Per kilogram cc	Per square meter cc	Amount cc	Per kilogram cc	Per square meter cc	
6/28/25	59.7	1.60	68	-6	3.9	36.8	2,570	43.0	1,605	4,065	68.0	2,540	Now taking 4½ grains thyroid daily omit July 4
7/ 8/25	61.2	1.61	64	-8	4.1	36.6	2,360	38.6	1,465	3,725	60.9	2,315	
7/12/25	61.0	1.61	61	-15	4.0	37.1	2,400	39.4	1,490	3,820	62.7	2,370	
7/16/25	61.7	1.62	61	-18	3.9	38.9	2,270	36.8	1,400	3,710	60.1	2,290	
7/19/25	61.7	1.62	58	-18	4.5	39.2	2,235	36.2	1,380	3,680	59.6	2,275	
7/28/25	63.6	1.64	60	-21	4.0	36.6	2,395	37.6	1,460	3,780	59.4	2,305	
8/ 2/25	63.5	1.64	52	-24	4.1	38.5	2,120	33.4	1,290	3,450	54.4	2,100	
8/ 6/25	63.3	1.63	55	-23	3.8	38.0	2,115	33.4	1,300	3,410	53.9	2,095	
8/ 8/25	64.8	1.66		-23	4.1	39.8	2,115	32.6	1,275	3,510	54.1	2,115	Started on 4½ grains thyroid daily
8/10/25	64.6	1.66	59	-22	4.2	36.9	2,210	34.2	1,330	3,500	54.2	2,105	
8/11/25	64.0	1.65	62	-18	3.9	35.6	2,300	36.0	1,395	3,575	55.9	2,165	Increased to 6 grains daily
8/13/25	63.6	1.64	58	-16	3.7	34.8	2,405	37.8	1,470	3,690	58.0	2,255	
8/16/25	63.3	1.63	74	-3	3.4	33.5	2,645	41.9	1,620	3,970	62.8	2,430	Decreased to 4½ grains daily
8/22/25	62.6	1.63	69	+3	3.4	30.8	2,720	43.5	1,670	3,930	62.8	2,410	Last few days alternating on 3 and 4½ grains daily
9/ 8/25	62.9	1.63	70	0	3.1	29.8	2,655	42.2	1,625	3,785	60.3	2,320	Reduced thyroid to 3 grains daily
9/19/25	64.0	1.65	66	-3	3.3	31.6	2,665	41.7	1,615	3,900	61.0	2,360	Ordered thyroid increased to 6 grains daily September 29

TABLE 9
Case of *myxædema* no 7
I female, age 42 years, height 165 cm

Date	Weight kg	Body surface sq m	Pulse	B M R. per cent	R B C. mill lions	Hematocrit per cent cells	Plasma volume			Total blood volume			Remarks
							Amount cc	Per kilogram cc	Per square meter cc	Amount cc	Per kilogram cc	Per square meter cc	
5/ 5/25	19.4	1.52	44	-10	4	36	62,295	46	51,510	3,620	73	52,360	Thyroid omitted April 25
5/ 9/25	49.4	1.52	51	-20	4	38	42,200	41	61,450	3,570	72	52,360	
5/14/25	48.9	1.52	57	-19	4	40	02,060	42	21,355	3,435	70	42,260	
5/15/25	49.7	1.54	60	-11	3	39	42,140	43	01,390	3,530	71	02,290	Started on 6 grains thyroid daily
5/23/25	49.4	1.52	75	-6	4	38	22,370	48	01,560	3,840	77	92,525	
5/30/25	48.5	1.52	62	-3	4	38	22,470	51	01,625	4,000	82	52,630	Ordered to increase thyroid to 9 grains daily June 3
6/10/25	47.1	1.50	70	+5	1	35	62,530	53	51,685	3,935	83	12,620	
6/22/25	48.0	1.51	73	-3	4	35	32,490	51	91,650	3,850	80	32,550	Increased thyroid to 12 grains daily
7/ 1/25	48.4	1.51	80	+5	4	34	82,625	54	31,730	4,025	83	32,650	Decreased to 7½ grains daily
7/13/25	48.5	1.52	75	+2	1	35	12,530	52	21,665	3,900	80	52,565	
7/22/25	47.3	1.50	71	+5	4	35	32,535	53	61,690	3,920	83	02,615	Decreased to 4½ grains daily
8/ 5/25	48.2	1.51	67	-1	4	36	12,470	51	31,635	3,870	80	42,560	Increased to 6 grains daily
8/14/25	18.7	1.52	68	-1	4	37	22,150	50	31,615	3,900	80	12,565	Ordered to increase thyroid to 12 grains daily September 4
9/ 9/25	49.9	1.54	68	+1	4	36	82,565	52	01,680	4,100	82	22,665	Decreased to 6 grains daily
9/19/25	48.8	1.52	66	+8	4	36	12,315	48	11,545	3,800	77	92,500	Decreased to 4½ grains daily and increased to 6 grains daily, October 3
10/ 5/25	19.8	1.51	70	+6	4	37	52,480	19	91,610	3,970	79	62,580	Decreased to 4½ grains daily
10/20/25	19.9	1.51	62	+6	6	40	22,180	13	71,415	3,650	73	22,370	
11/18/25	49.5	1.53	68	-1	4	40	12,275	46	01,490	3,800	76	82,480	
12/17/25	18.9	1.52	73	-3	4	38	82,360	48	11,555	3,855	79	02,540	

TABLE 8
Case of myxedema no 6
Female, age 52 years, height 159 cm

Date	Weight kg	Body surface sq m	Pulse	B M R per cent	R B C mil lions	Hematocrit per cent cells	Plasma volume			Total blood volume			Remarks
							Amount cc	Per kilogram cc	Per square meter cc	Amount cc	Per kilogram cc	Per square meter cc	
7/29/25	84.6	1.87	76	-24	4.4	45	62,325	27.5	1,245	4,285	50	62,295	Has been off thyroid two months
7/30/25	84.7	1.87	73	-30	4.6	43	12,400	28.3	1,285	4,225	49	92,260	
8/3/25	85.0	1.87	73	-30	3.7	42	12,570	30.2	1,375	4,440	52	22,375	
8/5/25	85.3	1.87	68	-26	4.0	40	12,460	28.8	1,315	4,110	48	22,200	
8/7/25	84.8	1.87	66	-30	4.2	41	02,535	29.9	1,355	4,300	50	72,300	Started on thyroid 6 grains daily Thyroid 19½ grains to date
8/10/25	83.5	1.86	84	-14	3.4	36	62,880	34.5	1,550	4,550	54	52,445	
8/11/25	82.7	1.85	91	-12	3.6	36	22,905	35.2	1,570	4,550	55	02,460	Reduced thyroid to 4½ grains daily
8/13/25	82.8	1.85	93	-7	3.6	37	12,810	34.0	1,520	4,475	54	02,420	
8/17/25	81.1	1.83	100	+8	3.7	36	72,910	35.9	1,590	4,600	56	72,510	Reduced thyroid to 3 grains daily
8/22/25	78.8	1.82	91	+7	4.1	39	22,725	34.6	1,500	4,480	57	02,470	
9/12/25	76.9	1.80	106	+15		39	52,890	37.6	1,605	4,780	62	22,655	Reduced thyroid to 1½ grains daily
9/22/25	76.7	1.80	101	+7	3.9	41	72,740	35.7	1,520	4,700	61	32,610	
10/6/25	76.5	1.80	93	+11	4.4	41	42,730	35.7	1,515	4,650	60	82,580	Increased thyroid to 4½ grains daily
10/27/25	75.9	1.79	111	+15	4.3	42	52,850	37.6	1,590	4,955	65	42,775	
11/25/25	74.6	1.78	95	+11	4.6	43	82,735	36.6	1,535	4,860	65	22,735	Increased thyroid to 6 grains daily
12/28/25	77.4	1.80	98	+1	4.4	42	62,800	36.2	1,555	4,880	63	22,715	
1/4/26	76.3	1.79	108	+11	4.5	42	52,915	38.2	1,630	5,070	66	52,830	Reduced thyroid to 1½ grains daily
1/12/26	75.5	1.79	103	+23	4.6	42	02,730	36.2	1,525	4,705	62	42,630	
1/19/26	75.6	1.79	115	+29	4.5	43	02,820	37.3	1,575	4,950	65	52,765	

TABLE 11

Case of myxedema no 9

1 female, age 39 years, height 157 cm

Date	Weight kg	Body surface sq dr	Pulse	B.M.R. per cent	R.B.C. millions	Hematocrit per cent cells	Plasma volume			Total blood volume			Remarks
							Amount cc	Per kilogram cc	Per square meter cc	Amount cc	Per kilogram cc	Per square meter cc	
5/18/25	55.1	1.54	63	-9	4.5	42	11,090	36	11,290	3,440	62	42,230	thyroid omitted May 10 daily Was then on 6 grams
5/26/25	55.8	1.55	54	-23	4.9	45	81,680	30	11,085	3,100	55	52,000	Began 2 mg oral thyroxin daily
6/2/25	56.0	1.55	57	-32	1.4	45	71,790	32	01,155	3,300	59	02,125	June 10, thyroxin 6 mg
6/10/25	56.0	1.55	57	-31	5.2	43	31,925	34	41,210	3,400	60	82,190	June 11, thyroxin 8 mg June 12 to 14, thyroxin 12 mg daily thyroxin 10 mg 10 mg thyroxin daily since June 15
6/24/25	55.3	1.54	61	-28	4.7	45	01,905	34	51,240	3,460	62	62,245	20 mg thyroxin intravenously
6/21/25	54.2	1.53	95	-6	4.5	45	01,910	35	81,270	3,530	65	12,310	
6/25/25	53.0	1.52	97	-3	4.9	44	42,035	38	41,340	3,660	69	02,410	
6/29/25	53.0	1.52	77	-4	4.2	38	22,300	43	41,510	3,725	70	32,455	
7/2/25	52.5	1.52	66	-11	4.4	40	52,220	42	31,460	3,730	71	02,455	
7/6/25	52.0	1.51	69	-12	4.6	42	61,935	37	21,280	3,360	65	02,240	
7/13/25	53.0	1.52	63	-22	4.2	42	61,840	34	71,210	3,210	60	52,110	Started on 7½ grams thyroxin daily
7/17/25	53.2	1.52	70	-15	4.0	39	52,180	41	01,435	3,600	67	72,370	
7/21/25	52.6	1.52	79	-5	4.5	41	71,970	37	41,295	3,385	64	42,230	Reduced thyroid to 6 grams daily
7/30/25	52.1	1.51	72	-6	3.8	41	41,935	37	01,280	3,300	63	12,180	Increased thyroid to 7½ grams daily
9/30/25	52.3	1.51	77	-14	4.7	41	22,065	39	51,370	3,520	67	32,330	
10/22/25	52.7	1.52	74	-5	4.0	40	02,075	39	41,365	3,460	65	72,280	

TABLE 10
Case of myxedema no 8
 Female, age 61 years, height 161 cm

Date	Weight kg	Body surface sq m	Pulse	B M R per cent	R B C mil- lions	Hematocrit per cent cells	Plasma volume			Total blood volume			Remarks
							Amount cc	Per kilogram cc	Per square meter cc	Amount cc	Per kilogram cc	Per square meter cc	
5/14/25	90.3	1.94	61	-15	4.9	43	92,950	32.7	1,520	5,250	58	22,705	Had been off thyroid 3 to 4 weeks
5/20/25	91.6	1.95	55	-21	5.0	44	62,670	29.1	1,370	4,825	52	62,475	
5/25/25	91.2	1.94	56	-23	5.0	45	82,405	26.4	1,240	4,445	48	72,290	
5/27/25	91.6	1.95	54	-26	5.2	46	42,620	28.6	1,345	4,885	53	32,505	
6/ 1/25	92.1	1.95	56	-25	4.4	43	22,790	30.3	1,430	4,915	53	42,520	Started on oral thyrovin, 2 mg daily Increased thyrovin to 4 mg daily Increased to 8 mg daily
6/ 5/25	91.7	1.95	60	-24	4.3	39	33,090	33.7	1,585	5,100	55	62,615	
6/ 8/25	91.0	1.95	57	-23	4.2	43	02,770	30.4	1,420	4,860	53	52,490	
6/12/25	91.0	1.95	54	-21	4.2	42	02,720	29.9	1,395	4,690	51	52,405	Started on thyroid extract June 12, 1 gram June 13, 9 grams June 14, 9 grams June 15, 7 grams June 16 to 18, 6 grams daily
6/18/25	88.4	1.92	79	-3	3.9	39	83,090	35.0	1,610	5,135	58	22,670	Reduced thyrovd to 5 grams daily
6/26/25	87.1	1.91	70	0	4.2	39	33,100	35.6	1,625	5,100	58	52,670	Reduced thyrovd to 4 grams daily June 24
7/ 7/25	84.4	1.88	77	-2	4.4	40	02,915	34.6	1,550	4,855	57	62,580	Increased thyrovd to 4½ grams daily
7/15/25	83.0	1.87	71	-2	4.5	38	93,010	36.3	1,610	4,925	59	42,635	
8/ 4/25	83.0	1.87	73	+3	4.0	38	93,100	37.4	1,660	5,070	51	22,715	
8/20/25	82.7	1.87	70	+6	4.1	38	03,315	40.0	1,775	5,350	64	72,860	
9/25/25	82.5	1.87	66	+4	4.3	38	83,160	38.3	1,690	5,165	62	72,760	
10/ 9/25	82.9	1.87	66	+7	4.1	40	62,910	35.2	1,555	4,000	59	22,620	
11/11/25	82.7	1.87	79	-2	4.4	39	93,410	41.2	1,825	5,675	68	53,035	

the level was as low in ten days as it was in the following ten while in patient number 5 it kept slowly dropping over a period of 5 weeks and, if giving thyroid extract had been withheld longer might have dropped further. Plummer and Boothby (22) have estimated that it takes fifty to sixty days for the effects of a single intravenous injection of thyroxin to wear off. Five of these patients were without thyroid extract for considerably shorter periods than this, it is quite possible that the changes in the plasma would have been more marked had the administration been withheld for a longer time.

Cases previously untreated might show greater changes, as case 2 illustrates. When first seen, she presented the most marked symptoms of myxedema I have ever seen. Just after she had started 9 grains of thyroid extract daily and while her metabolic rate was still low, the total plasma was 1850 cc. Her veins were small, difficult to distend and puncture, her blood was extremely viscous and clotted rapidly. The day following the first plasma volume determination, her basal metabolism was -19 per cent. Two days afterward the total plasma was 2350 cc. The fact that she took 9 grains of thyroid extract daily for several days before the first estimation of plasma volume indicates that this value may have been wrong, that is to say too low. The second estimation two days later, however, as has been said was less than that subsequently found even after the non-administration of thyroid extract for six weeks, after treatment with it for six months. Under these circumstances the first observation then apparently too low may have been approximately correct. If so, subsequent determinations represent an increase in plasma volume of at least 62 per cent over the myxedematous level.

VI RELATION BETWEEN PLASMA VOLUME AND BASAL METABOLIC RATE

A parallelism appears to exist between plasma volume and basal metabolism (figs 1, 2 and 3 plotted from the data in tables 6, 7 and 10).

Inasmuch as basal metabolism is a function of surface area, a comparison with plasma per square meter is justified. In interpreting such charts, it must be born in mind that there is a 5 per cent error in both plasma volume and basal metabolic rate determinations and that, therefore, only the general trend of the curves is of significance.

Patients may show a marked plasma volume decrease on omission of thyroid extract, with practically no change in weight. This is well illustrated by case 4.

The percentage increase in total plasma over the myxedematous level varied from 14.0 to 35.0 per cent, that in the plasma per kilogram from 16.2 to 41.7 per cent, and that in the plasma per square meter from 17.1 to 37.7 per cent. The average increases in the three for all nine patients were 22.9, 28.5 and 25.2 per cent, respectively. With an average basal metabolism increase over the myxedematous level of 38 per cent the last seven cases showed average percentage increases in total plasma, plasma per kilogram and plasma per square meter of 23.2, 28.9 and 25.2 per cent, respectively.

I have ascertained the average plasma volume and basal metabolism figures for each patient both while off and on the administration of thyroid extract, as well as the gross and percentage increases in the two over the myxedematous level (table 2) as well as the data collected on each individual patient, the order of patients in table 2 being preserved throughout (tables 3 to 11).

To prove that the plasma volume increase endures as long as treatment is continued and is not dependent on the elimination of excess fluid by diuresis, a study was made of all of the patients in whom a control observation was made before omitting the administration of thyroid extract, in all of them it was found that a decrease was observed after the omission. Two of these patients (cases 4 and 5) had been treated for 14 and 15 months respectively, but in both the omission produced a well marked decrease in plasma volume and subsequent administration, a return to the previous level. All patients (except number 3) were followed furthermore, for several months after treatment was begun and in all the increase in plasma volume persisted.

V TIME REQUIRED FOR PLASMA VOLUME REDUCTION TO OCCUR AFTER OMISSION OF THYROID EXTRACT

The time required for plasma volume reduction to occur after omitting the administration of thyroid extract varies considerably from individual to individual. In all of the cases a well marked change had occurred within ten days after omission. In patient number 4

At the suggestion of Dr Means, these data of the seven cases have been plotted (fig 4) For each patient represented in figure 4, a point was plotted for the average without thyroid extract administration and another for the average with treatment, these points have been connected with a straight line (fig 5)

This figure gives at first the impression of a striking parallelism between the seven lines When inspected closely, however, it will

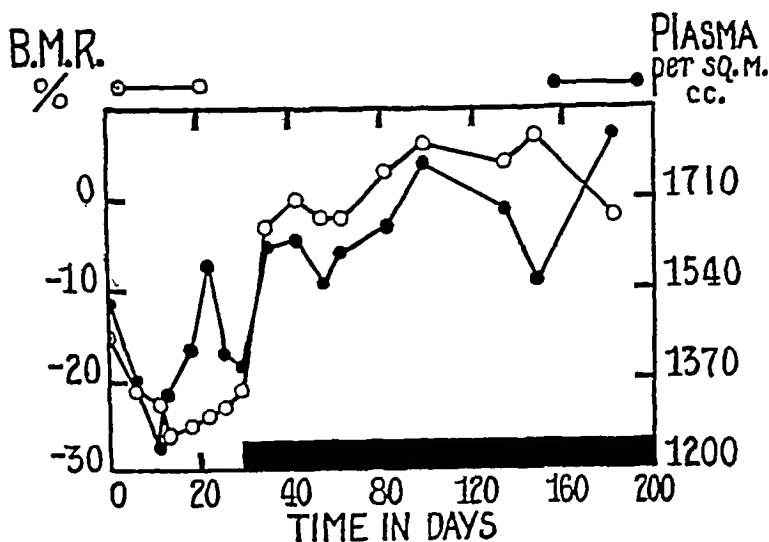


FIG 3 CASE OF MYXEDEMA NO 8

These curves likewise show parallelism between B M R and plasma per square meter Thyroid extract administration omitted about three weeks before the first recorded observation

be observed that there are differences in the reactions of different patients These are related neither to the differences in basal metabolic rate nor to those in maintenance dosage of thyroid extract They are probably manifestations of individual variation and represent fundamental differences in human constitution They may be likened to the marked variations in the extent of anhydremia produced by the same dose of insulin in different rabbits or approximately the

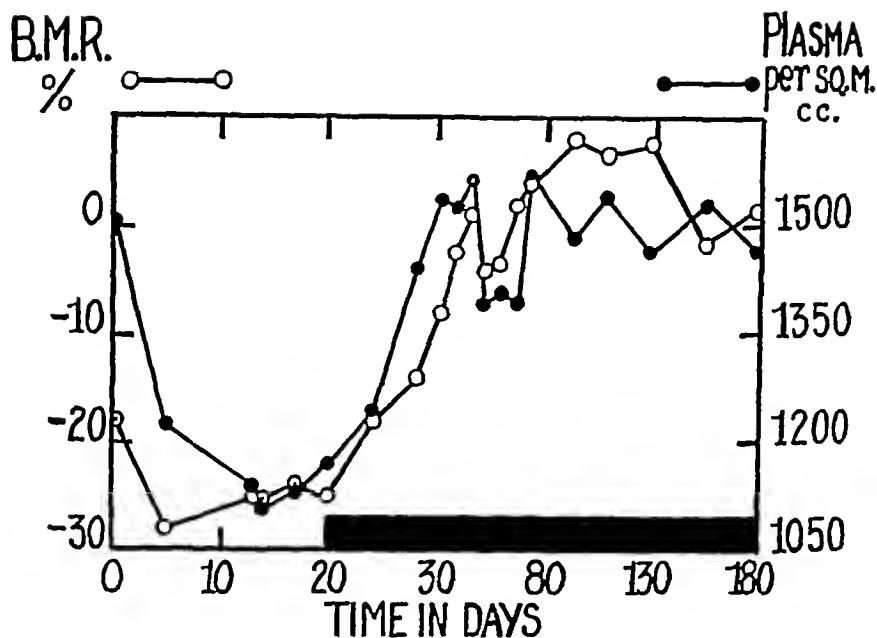


FIG 1 CASE OF MYXEDEMA NO 4

Black area in this and subsequent figures denotes the period of thyroid medication. The curves show striking parallelism between basal metabolic rate (B.M.R.) and plasma per square meter. Thyroid extract administration omitted at beginning of chart.

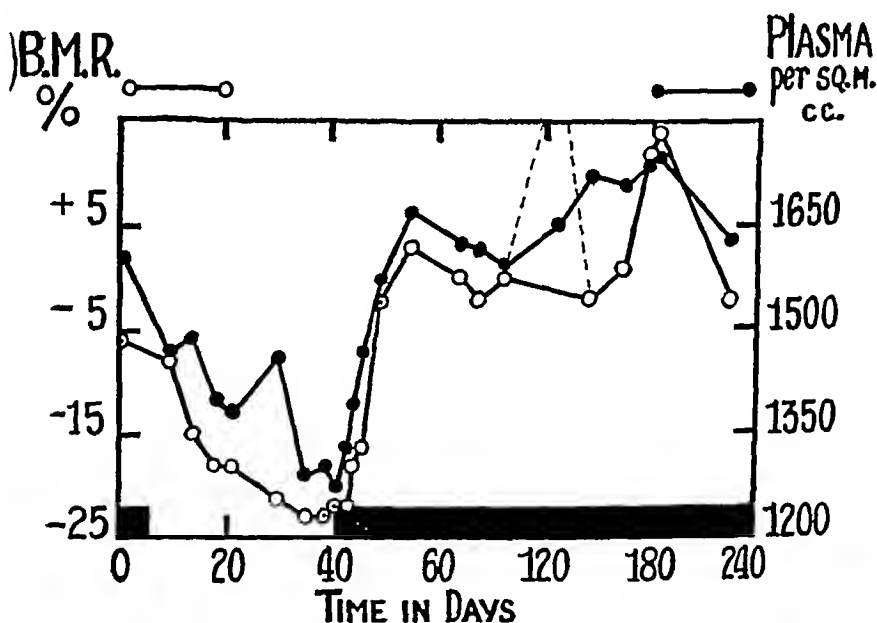


FIG 2 CASE OF MYXEDEMA NO 5

These curves also show parallelism between B.M.R. and plasma per square meter. Note the slow fall on omission of thyroid extract therapy and rapid rise on administering it. Inasmuch as several B.M.R. observations on 6 grains thyroid daily ranged from -3 per cent to +3 per cent, one observation of +22 per cent on this dose is undoubtedly high and is connected to the others by dotted lines.

tain the normal basal metabolic level in patients with myxedema who show about the same reduction in basal metabolism ⁴

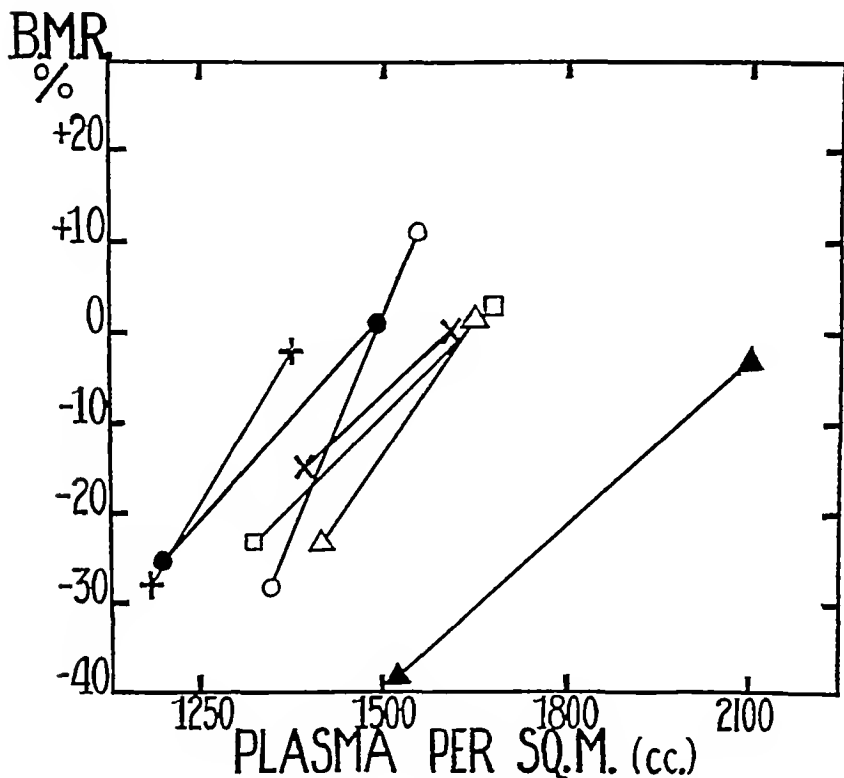


FIG 5 THE RELATION OF B M R TO PLASMA PER SQUARE METER IS SHOWN IN ALL THE CASES

For each case represented in figure 4, a mark has been plotted for the average B M R.—plasma per square meter point when free from, and another during, thyroid extract administration, the two are connected by a straight line

⁴ It is not implied that basal metabolism and plasma volume always increase together. There are no doubt different types of accelerated and diminished cellular activity. For example, the oxygen consumption is increased in fever, but evidence thus far collected suggests that the blood is concentrated.

same weight (Drabkin and Edwards (23)), to the marked variation in the severity of myxedematous symptoms in different patients with essentially the same reduction in basal metabolic rate and to the marked variation in the amount of thyroid substance required to main-

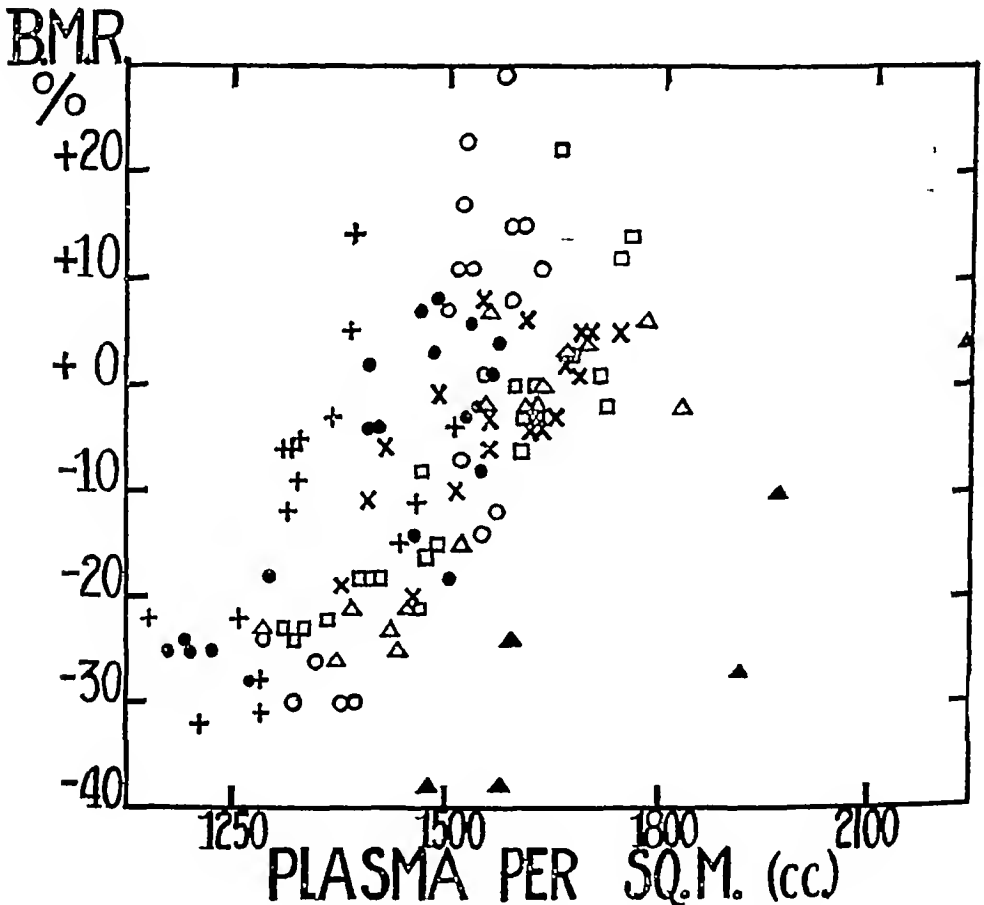


FIG 4 B M R vs PLASMA PER SQUARE METER ALL CASES

Observations on case 3 shown by black triangles
 Observations on case 4 shown by dots
 Observations on case 5 shown by squares
 Observations on case 6 shown by circles
 Observations on case 7 shown by crosses
 Observations on case 8 shown by white triangles
 Observations on case 9 shown by pluses

four times the maintenance dose produce only slightly greater increases in plasma volume and basal metabolism than the maintenance dose (figs 6 and 7) One observes the abrupt change in direction of the two lines when the dose is pushed beyond the maintenance dose

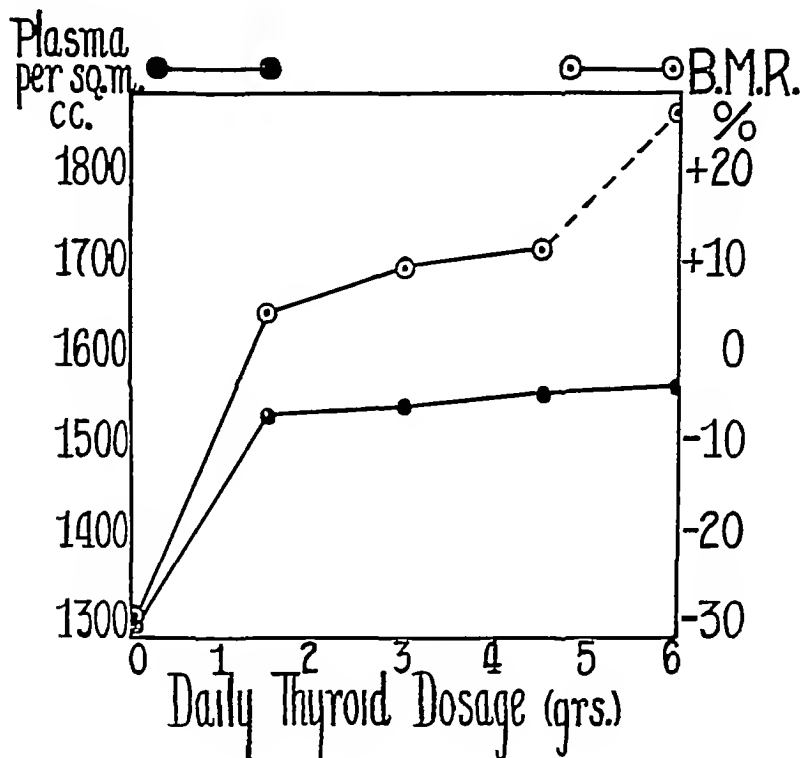


FIG 7 CASE OF MYXEDEMA No 6

These curves likewise show the effect on B M R and plasma volume of increasing dose of thyroid extract beyond the maintenance dose. The B M R on 6 grains daily represents only one observation and is probably high inasmuch as one of the three B M R observations on $4\frac{1}{2}$ grains daily was -23 per cent.

Yet, in spite of little change in plasma volume or basal metabolism, these patients would often complain of precordial pain, palpitation epigastric cramps and dysmenorrhea. One woman (case 7) whose

VII THE RELATION BETWEEN THE DOSE OF THYROID EXTRACT⁵ AND THE DEGREE OF PLASMA INCREASE IN THE SAME INDIVIDUAL

It is striking that once the basal metabolism has been restored to its normal level, increases in the dose of thyroid extract up to three and

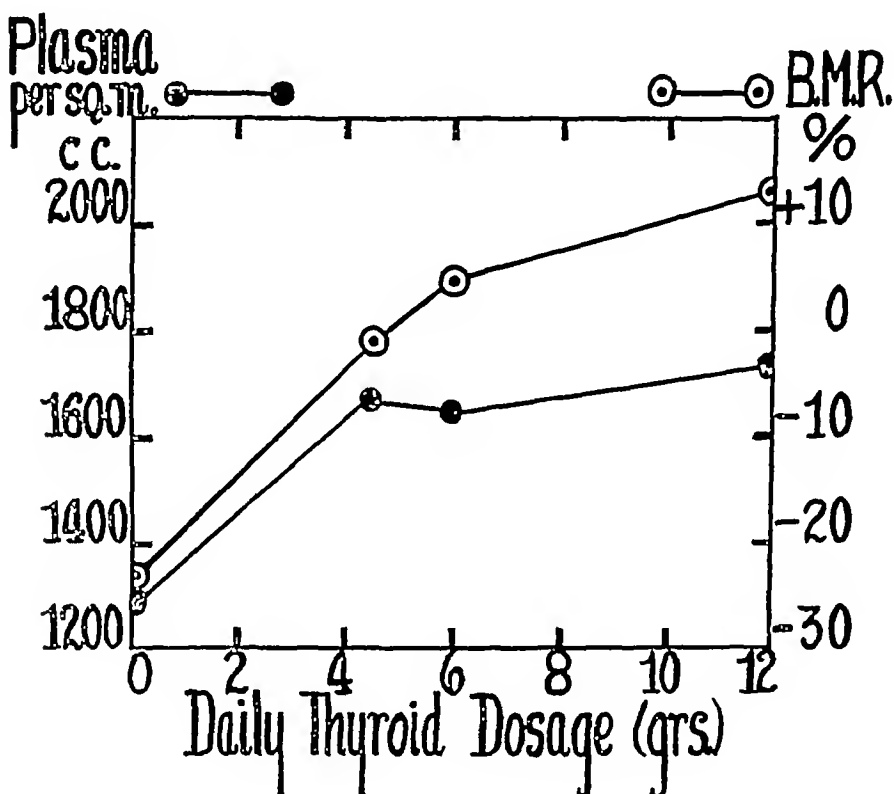


FIG 6 CASE OF MYXEDEMA No 5

These curves show the effect on B M R and plasma volume of increasing the dose of thyroid extract beyond the maintenance dose. Each point represents the average of at least two observations.

⁵ Burroughs Wellcome and Company's thyroid extract was used throughout this work, except for the first four weeks in case 8, during which Armour's 1 grain tablets were used. In the last six patients Burroughs Wellcome and Company's thyroid preparation was used in $1\frac{1}{2}$ grain tablet strength only. In cases 8 and 9 oral thyroxin in large doses was used without effect on basal metabolic rate or plasma volume, whereas thyroid extract subsequently produced a prompt and sustained increase in both. Twenty milligrams of thyroxin were injected intravenously in case 9, while she was myxedematous. A well marked increase in basal metabolic rate and plasma volume occurred, the data being recorded in table 11.

TABLE 12
Effect of thyroid extract administration on basal metabolic rate and plasma volume in a normal individual
 Female, age 36 years, height 152 cm

Date	Weight	Body surface	Iube	B M R	K B C	Hmato- crit	Plasma volume				Remarks
							Amount	1 cr kilo- gram	1 cr square metr		
									cc	cc	
1/18/25	60.9	1.57	60	-11	4.6	45.7	2,210	36.2	1.410	April 16, thy roid 3 grams April 17, thy roid 1½ grams April 20, thy roid 1½ grams Started on thy roid 9 grams	
4/21/25	62.0	1.59			4.6	44.7	2,210	35.6	1,390	Decreased to 3 grams daily April 25 Increased to 6 grams daily	
5/ 4/25	62.1	1.59	64	-10	1.5	41.5	2,125	34.2	1,335		
5/12/25	64.6	1.61	95	-2	4.6	41.7	2,515	39.4	1,540		
5/13/25	63.7	1.61	76	0	4.3	42.5	2,500	39.2	1,550		
5/16/25	62.3	1.59	79	-1	1.3	43.5	2,170	34.8	1,365	Decreased to 1½ grams daily	
6/ 8/25	64.8	1.62	79	-1	1.3	38.5	2,540	39.2	1,570	Increased to 6 grams daily	
6/10/25	64.8	1.62	72	-2	4.0	38.5	2,150	37.8	1,510	Thy roid omitted	
6/15/25	63.9	1.61	62	-15	1.6	40.1	2,315	36.8	1,460		
6/19/25	62.2	1.59	59	-12	1.5	41.0	2,250	36.2	1,415		

auricles were fibrillating and whose maintenance requirement was $7\frac{1}{2}$ grains daily had an attack of unconsciousness (probably Stokes-Adams)⁶ on two occasions when the dose was increased to 12 grains daily. She had had none before and has had none since.

One may compare the difficulty experienced in raising basal metabolism and increasing plasma volume once the basal metabolism is normal with the difficulty encountered in raising both in a normal individual by giving thyroid extract (table 12). This individual probably belonged to the so-called "low rate" group of patients. She showed only a slight increase in basal metabolism and a suggestive slight increase in plasma volume on taking $4\frac{1}{2}$ to 6 grains of thyroid extract daily. Yet she had precordial pain, palpitation, epigastric pain and severe dysmenorrhea, all of which disappeared when the administration was stopped.

Data were collected also from a postoperative exophthalmic goitre patient who may be regarded as approximately normal (table 13). The data begin to be recorded about four months after a subtotal thyroidectomy, that is to say, after the weight had become stationary. Iodine (Lugol's solution) was first given for about $5\frac{1}{2}$ weeks. The basal metabolism on iodine treatment dropped slightly to a stationary level around -5 per cent. Thyroid extract was then administered in slowly increasing doses over a period of about $2\frac{1}{2}$ months. The patient was given $13\frac{1}{2}$ grains daily for approximately one and one-half months. There occurred a slight increase in basal metabolism (18 points) and possibly a slight increase in plasma volume (1560 cc per square meter to 1670 cc per square meter). The patient experienced no toxic symptoms on $13\frac{1}{2}$ grains of thyroid daily, a dose sufficiently large to produce intoxication in most patients with myxedema. These last two patients illustrate what appears to be a matter of importance, namely, variations in the amount of thyroid extract re-

⁶ It is of interest in this connection that Willius (48) recommends the use of small doses of thyroid extract (1 to 2 grains daily) in the treatment of the Adams-Stokes syndrome and that Aub and Stern (49) report that the daily administration of 28 grains of Burroughs Wellcome and Company's thyroid extract for several weeks produced no cardiac symptoms in a patient with complete heart block, in spite of an increase in basal metabolism to +47 per cent.

quired to produce intoxication in different normal individuals⁷ It is significant that when one plots the basal metabolism and plasma per square meter against the dose of thyroid extract for the patient after thyroid operation (table 13, fig 8) one gets the same type of curves as is seen in figures 6 and 7 after the maintenance dose of thyroid is reached

Other cases could be cited These are sufficient, however, to illustrate that once a patient with myxedema has been given sufficient

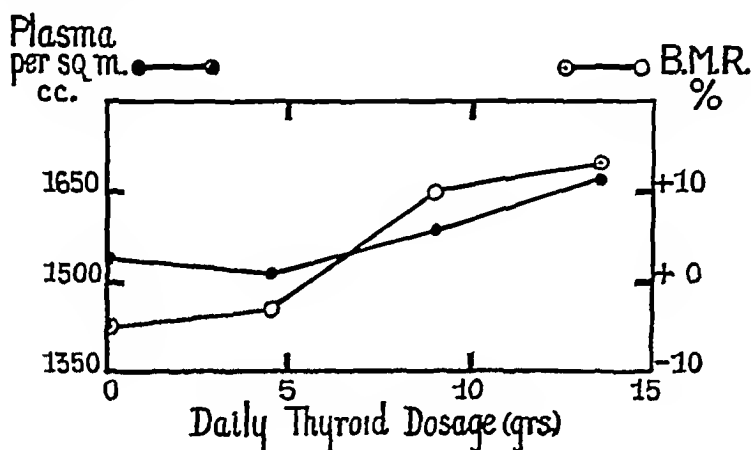


FIG 8

These Curves present the record of a case of exophthalmic goitre during a period of thyroid extract therapy beginning four months after a subtotal thyroidectomy (Table 13)

Note that the abrupt initial rise (maintenance dose) seen in figures 6 and 7 is lacking

thyroid to maintain a normal basal metabolism and plasma volume, one is dealing with an essentially normal individual When the dose of thyroid extract is further increased up to two or four times the maintenance dose, the increases in basal metabolism and plasma vol-

⁷ It is realized, of course, that post operative patients with exophthalmic goitres, although not grossly myxedematous, may possibly require more thyroid extract before they become toxic than normal individuals This may account in some measure for the difference, but probably not completely

TABLE 13
Effect of thyroid extract administration on plasma volume and basal metabolism in a post-operative exophthalmic goitre
 Female, age 37 years, height 152 cm

Date	Weight kg	Body surface sq m	Pulse	B M R per cent	R B C millions	Hemato- crit per cent cells	Plasma volume			Remarks
							Amount cc	Per kilo- gram cc.	Per square meter cc	
11/23/25	49.9	1.45	62	+5	4.0	39.3	2,280	45.7	1,570	Started on KI (saturated solution), 5 drops t i d
12/ 7/25	50.8	1.46	69	-5	4.3	41.5	2,340	46.1	1,600	Started on thyroid extract 4½ grains daily Reduced KI to 5 drops daily
12/31/25	51.4	1.46	60	-4	4.4	41.6	2,210	43.0	1,515	
1/11/26	51.0	1.46	66	+1	4.2	40.9	2,160	42.4	1,480	Increased thyroid to 9 grains daily
1/18/26	50.9	1.46	64	-7	3.9	40.4	2,260	44.5	1,550	
1/27/26	50.1	1.45	78	+6	4.1	40.0	2,280	45.5	1,570	Increased thyroid to 13½ grains daily
2/ 2/26	50.0	1.45	76	+10	4.4	40.4	2,260	45.2	1,560	
2/12/26	49.9	1.45	80	+15	4.0	39.6	2,360	47.4	1,630	
2/18/26	49.6	1.44	81	+9	4.0	39.6	2,340	47.2	1,625	
2/26/26	49.7	1.44	83	+17	4.4	40.4	2,405	48.5	1,670	
3/17/26	48.4	1.42	80	+13	4.1	39.9	2,440	50.5	1,720	

seven months and was then omitted for about six weeks (May 14, 1924, to June 27, 1924). During this period the average plasma volume was 2615 cc. Observations made from July 3, 1924, to September 29, 1924, while taking thyroid extract show an average hematocrit value of 35.6, an average red count of 3.9 millions and an average blood volume of 4600 cc. Let it be assumed that the plasma volume in the myxedematous state before she received thyroid extract was the same as it was when its administration was omitted, namely, 2615 cc. This, with an hematocrit value of 26.5 gives a total blood volume of 3560 cc., or 1140 cc. less than the total volume after she had reached an equilibrium on thyroid extract treatment. This seems a conservative estimate, since the first two plasma volume observations of 1850 and 2350 cc. (although questionable, as stated on page 497) would give still lower total blood volumes for her original myxedematous condition.

Case 2 (table 4) presents a second example. It was possible to withhold thyroid extract from this patient for a period of over three months (January 7, 1924, to April 16, 1924). It will be observed that although the hematocrit reading increased at first, towards the end of the period it decreased slightly. Then after medication began again (April 16, 1924) there occurred following a further decrease, an increase which set in about one month later and gradually became more marked during the following four months. If the average of the last three total blood volumes (March 22, 1924, to April 14, 1924) when he was not taking thyroid extract be compared with the average of the last five (August 14, 1924, to September 11, 1924) during its administration, an increase in total blood volume of 1245 cc. will be observed. The data suggest that the original cell volume was less than when treatment was subsequently omitted, considering that the red count on admission averaged between 2.0 and 3.0 millions as compared with 4.0 millions during the untreated period beginning January 7, 1924. The change of 1245 cc. in total volume is one actually observed and is probably less than the change that would have been recorded had observations been made before thyroid therapy was begun.

Case 6 had been without thyroid extract for two months when last admitted to the hospital. The total volume (5 observations made

ume are proportionately much smaller than those produced by the maintenance dose and are of the same magnitude as the changes produced in both by giving thyroid extract to a normal individual. This phenomenon is probably fundamental and seems to be the result of calling into play some defense mechanism, some one of the body's factors of safety to deal with excess.

VIII CHANGES IN TOTAL BLOOD VOLUME

In untreated cases of myxedema a well marked secondary anemia with a hemoglobin of about 60 per cent and a red blood cell count of between 3,000,000 and 4,000,000 is the rule rather than the exception and has been emphasized by numerous observers including Minot (24), Emery (25), and recently McKunde (26). She found a decrease in hematocrit values in experimental myxedema in rabbits from 30 to 31 per cent to 23 to 24 per cent, and a reduction in hemoglobin of 30 to 40 per cent. This reduction in hematocrit value does not represent the total reduction in volume of circulating cells because there was probably also a reduction in plasma volume. Even if the plasma volume is concentrated only 15 per cent as the result of thyroidectomy, then an hematocrit reading of 23 really is too high and represents one of 19.5 based on the original quantity of plasma. If the original hematocrit value were 30 this would represent a reduction in volume of circulating cells of over one-third and in total blood volume of over one-fifth, reductions which may fairly be assumed.

The reduction in red count and cell volume following the omission of thyroid extract administration is a slow one, the first response is in fact an increase due to plasma reduction. It is manifestly unfair to keep patients with myxedema off the administration of thyroid extract long enough to allow the red count and cell volume to be lowered. There is evidence, however, in the data presented which allows one to draw justifiable conclusions about changes in total blood volume.

For example, consider again case 1 (table 3). Her hematocrit reading just after starting the administration of thyroid extract but before the drug had had any effect, averaged 26.5, with an average red count of 3.1 millions. Thyroid extract was administered for about

and Bock (27)) Bolton produced marked edema by reducing the lumen of the inferior vena cava to one-third in cats. Myxedema, on the other hand, probably has a much more complex mechanism as a result of changed osmotic relations between plasma and tissue cells. This subject is discussed later in greater detail.

In cardiac edema as the weight increases the total plasma increases, whereas in myxedema as the weight increases the total plasma decreases. This is illustrated in figures 9 and 10, where the weight curves and total plasma volumes are plotted for a case of cardiac edema and case 5 with myxedema. The data for figure 9 were collected from

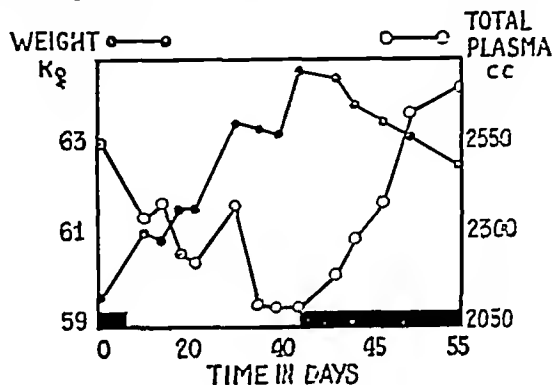


FIG. 10 CASE OF MYXEDEMA NO. 5

These curves show decrease in plasma volume with increase in weight and increase in plasma volume with decrease in weight in contrast to those in cardiac edema (fig. 9).

a case of chronic myocarditis which I was fortunate enough to observe while fully compensated, during the progress of edema and as edema was subsiding.⁸

Another point of interest in cardiac edema is that the osmotic

⁸ There is considerable evidence in the literature to support my findings in cardiac edema. As early as 1884 Oertel (29) taught that in cardiac edema, hydropic plethora was present. Stintzing and Gumprecht (30) found hydropic plethora not only during the height of dropsy, but also at the onset. Askanazy (31) concluded from his observations that the blood serum was always diluted, dilution depending upon the extent of the dropsy. Bolton (32) found a well marked increase in blood volume in animals in experimentally produced "dropsy."

from July 29, 1925, to August 7, 1925) averaged 4270 cc before the administration of thyroid extract was started August 7, 1925, and the 5 last observations, made in the fourth month after starting treatment averaged 4895 cc an increase of over 600 cc

From such data, together with experimental observations in animals following thyroidectomy, it appears that increases in total blood volume of one-fourth over the myxedematous level (i.e., 800 to 1000 cc of whole blood in human beings) are not uncommon

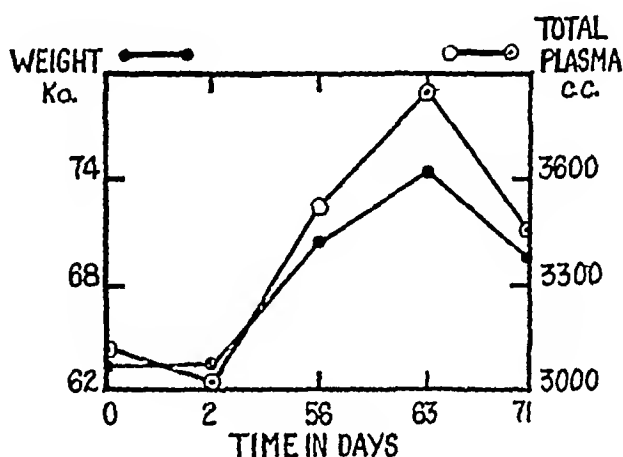


FIG 9 CARDIAC EDEMA

These curves show increase and decrease in total plasma volume with increase and decrease in edema (represented by weight curve)

IX A COMPARISON OF MYXEDEMA AND CARDIAC EDEMA

Myxedema and the edema of heart disease represent two totally different types of edema and, by comparing them, some light is thrown on the nature of both. In cardiac edema, the excess tissue fluid is chiefly intercellular and the skin pits on pressure. In myxedema the excess tissue fluid is chiefly intracellular and the skin does not pit on pressure. Cardiac edema may be explained on the basis of an increased capillary filtration pressure due to venous congestion (that is to say, this may be the underlying cause of all blood and tissue changes). Indeed, when one stands at the position of attention for as long as an hour, the cardiac output decreases, the venous pressure rises and a measurable increase occurs in the diameter of the calf (Field

injected Differences in the depth of injection are unavoidable Irritation of a sensitive skin often masks the end point I have, however several observations in four cases under and without the influence of thyroid extract and I present the averages in each case They consistently show a more prolonged disappearance time before or in the interval of thyroid omission than during the period of administration

Case number	Average without thyroid extract	Average on thyroid extract
	<i>minutes</i>	<i>minutes</i>
4	67	40
5	71	56
6	122	95
9	63	44

It is interesting to notice that in all other types of edema Aldrich and McClure found the disappearance time markedly diminished The wheals usually disappeared in 7 minutes or less over markedly edematous areas Although the time was usually greater in non-edematous than in edematous areas of the same patient, the time in non-edematous areas of edematous patients was usually much less than the normal of 50 or 50 plus minutes In a case of nephrosis at this hospital it was observed that in the legs where edema was marked the wheals disappeared in 1 to 2 minutes whereas in the arms where no pitting edema was present the wheals took 12 to 14 minutes to disappear Obviously this rapid disappearance in edema cannot be due to capillary absorption for the following reasons

1 The blood has a low osmotic pressure (as referred to protein) and can not properly absorb fluid from the tissues Yet the wheal often disappears at 40 to 50 times the normal rate

2 Differences in the chemical composition of blood sufficient to account for differences in disappearance time of 6 to 14 times are not known to exist

3 The disappearance time increases as edema subsides, a relation just the opposite of what one would expect if disappearance were due chiefly to absorption by the blood

One could account for the differences in disappearance time in myxedema before and after the administration of thyroid extract on the basis of changes in the rate of blood flow assuming that the dis-

pressure of the serum as referred to protein is low (Gavaerts (28)) whereas the data in myxedema thus far collected suggest that the serum protein is high. Inasmuch as hunger edema is also characterized by low serum protein and massive pitting edema, Oberndorfer's (1) finding of a markedly diminished weight of the thyroid gland in this condition appears to be of no consequence from the standpoint of etiology of the edema.

X THE EFFECT OF THYROID ADMINISTRATION ON THE PLASMA VOLUME IN CRETINS

A few observations on three cretins have been collected. Two of these (aged 4 and 5 years) showed practically no change after taking fairly large doses of thyroid extract for two to four weeks. The third case (aged 7 years) showed a 14 per cent increase, in total plasma after receiving 3 to 4 grains daily for three weeks. There are, however, not enough data to draw conclusions from. These findings are interesting in view of those of Hammett (50) that thyroidectomy in male rats 75 days of age produced much less increase in refractive index and much less reduction in water percentage of serum than it did in sexually mature male rats 100 days of age.

Young rabbits when thyroidectomized show reductions in hematocrit values which are less marked than those shown by mature rabbits (McKunde). An increase in hematocrit readings on giving thyroid extract has been observed in human cretinism (Talbot (33)).

XI THE RATE OF DISAPPEARANCE OF INTRADERMALLY INJECTED SALT SOLUTION IN MYXEDEMA BEFORE AND AFTER THYROID EXTRACT ADMINISTRATION

The Aldrich-McClure intradermal skin test (34, 35) was used. Sodium chloride solution (0.2 cc of an 0.8 per cent) was injected intradermally in two places about 2 cm apart on the flexor surface of each forearm a little below the bend of the elbow. The time at which an elevation ceased to be palpable on gently rubbing the finger over the point of injection was taken as the end point of the period of absorption. I do not consider the test of much value. Leakage, no matter how careful the technique, causes variation in the amount of fluid

In comparing myxedema with cardiac edema it was suggested that the increased tissue fluid in myxedema was probably for the most part intracellular. Tatum (38) has shown that in experimental cretinism in rabbits, while there is a varying amount of intercellular serous deposit, the greater changes are in the cells. He demonstrated that there were hydrophic changes in cells of the heart, liver, kidney, smooth and striated muscle. These experiments then give evidence to the effect that there may be increased water content of tissue cells.

That the water content of the tissues is increased in myxedema is a generally accepted fact. Recently McKunde has reported that the water content of dried fat-free tissue in thyroidectomized rabbits is higher than normal and that they lose weight and water when fed with thyroid extract. Hildebrandt has shown that the intravenous injection of thyroxin in rabbits causes not only a loss of water from the tissues but also of salt. It is, moreover, generally admitted that the secretion of the thyroid gland stimulates sodium chloride metabolism.

In myxedema there are then two phases in close association, first a reduction in the total quantity of circulating plasma, and second an increase in water, protein, and salt in the tissues. It is my opinion that both these phenomena are merely by-products of the fundamental change which thyroid insufficiency produces in the activity and chemical composition of tissue cells.

It is interesting in this connection to refer to the work of Barbour and Hamilton (39), (40), (41) who have demonstrated that cold produces blood concentration, and heat blood dilution, and that in cold anhydremia there is an increase in the water content of skin and muscle. They have not demonstrated whether this increased tissue water is intra- or extracellular. The idea is suggested, however, that cold and thyroid insufficiency may produce similar types of slowed cellular metabolism and that a by-product of both is probably an increased osmotic pressure of tissue cells.

Ellinger (42) believes that the influence of thyroid extract on the viscosity of blood serum is independent of the albumen content of the latter, and that in this respect its action is similar to that of caffeine and other diuretics.

Loeb (43) reports that Embden and his associates found that thyroid extract, when added to certain substances increases the rapidity

appearance in this case were chiefly the result of absorption by the blood. Until we know more precisely, however, what the mechanism of disappearance is, it is useless to say more on the subject. The results illustrate one more difference, nevertheless, between myxedema and all other types of edema. Pilcher (36) on injecting codein intradermally noticed that the injection wheals disappeared more rapidly than normal in a case of myxedema and in one cretin, as well as in other types of edema. It is important in such instances, however, not to confuse irritation with diffusion and absorption phenomena.

XII DISCUSSION

Magnus-Levy (37) showed that there was accumulation of albumen in the tissues in myxedema and thought that some of the increase was intercellular. Eppinger (11) assumed that this phenomenon was the cause of edema in myxedema by virtue of the affinity of albumen for water and salt. He inclined to the view, that in edema of this type intercellular albumen was a more important constituent than intracellular albumen and based this conclusion on the fact that when isotonic salt solution was injected subcutaneously in myxedematous dogs, protein became mixed with it whereas this did not occur in normal dogs. In his opinion protein admixture was responsible for slow absorption. Eppinger believed in fact that increased tissue albumen accounted for all types of edema. Boothby et al (51) confirmed the observation of Magnus-Levy and showed that the nitrogen-water ratio of the weight lost by one normal and by each of two myxedematous individuals as a result of intravenous thyroxin administration was 1.9, 2.0 and 1.9 per cent respectively. These authors, therefore, concluded that the edema of myxedema 'corresponds apparently to an increase in the reserve or deposit protein' and "is an albuminous colloid fluid with a nitrogen-water ratio higher than the average of 1.1 per cent for human blood serum and identical with that of egg white which contains approximately 2 per cent nitrogen, and definitely less than that of muscle which contains over 3 per cent." They emphasized the fact that on the contrary the nitrogen content of the edema of cardiac or renal origin is only 0.05 to 0.001 per cent, a negligible amount.

In brief, the reduction in plasma volume in myxedema is associated with an accumulation of albumen, water and salt in the tissues. It is a debatable point as to how much of this increase is intercellular and how much intracellular. Eppinger's finding of a protein admixture in subcutaneously injected isotonic salt solution in myxedematous dogs, suggests that a considerable part of the increase may be intercellular. On the contrary the contrast between myxedema and cardiac edema, together with Tatum's finding of an hydropic condition of various tissue cells in thyroidectomized rabbits, indicate that the larger part of the increase may be intracellular. A factor in these changes may be diminished permeability of cell membranes. The fundamental cause of all the blood changes however, would appear to be a decrease in the rate of metabolism of tissue cells, with a diminution in the demand for oxygen and carbon dioxide transport. The decreased quantity of circulating medium and the reduction in minute volume of the heart seem to be adaptations of the circulatory system to this diminished demand. I have suggested that the slowed cellular metabolism may increase cellular osmotic pressure, but the various steps in the process by means of which water is slowly withdrawn from plasma and stored up in the tissues, are yet to be worked out.

XIII SUMMARY AND CONCLUSIONS

In nine patients with myxedema the total plasma volume increased on the average 22.9 per cent, the plasma per kilogram 28.5 per cent, and the plasma per square meter 25.2 per cent, when given thyroid extract.

From observed and calculated increases in total blood volume, it is estimated that the administration of thyroid extract in myxedema not infrequently produces total blood volume increases of 25 per cent.

The plasma decrease on omission of thyroid extract and the increase on administering it, occur much more rapidly than the same changes in the volume of cells. Plasma reduction is usually well marked within ten days after omitting the drug.

A parallelism exists in myxedema between basal metabolism and plasma volume.

Once the basal metabolism has been restored to normal, further increases in the dose of thyroid extract up to four times the mainte-

with which the latter pass in vitro through a membrane of frog's skin. He also reports that "Recent experiments in Asher's laboratory have shown that under the influence of thyroid, methylene blue passes more rapidly from the body fluids into glandular structures in the membrane nictitans of the frog. Thyroid extract would thus appear to have an effect opposite to that of calcium chloride and adrenalin." In view of such reports one must admit that altered permeability of cell membranes may play a rôle in the effect of thyroid extract on fluid distribution in myxedema, but it would appear to be a minor rôle and to be explicable on the theory that cell membranes share in the general myxedematous condition.

Means (17) has well remarked that "However obscure the function of the endocrine glands may be, we can agree, I think, that they are all concerned in one way or another, direct or indirect, with metabolism.

If the secretions of endocrine glands influence cell activity, they must also influence the supply of the medium upon which such activity depends." We know that insulin produces a well marked anhydremia, that adrenalin causes some anhydremia and that pituitrin has a marked effect on water elimination. Krogh (44) indeed thinks that it decreases capillary permeability.

Means also referred to the observation of Bock and Field (45) that in myxedema the administration of thyroid extract markedly increased the minute volume of the heart, initially very low. He referred to the reduction in the volume of blood and in cardiac output as manifestations of the variation in the supply of circulating medium with the demand. Daly (46) has shown that mere increases in the volume of blood, other things being equal, increase the cardiac output. As already stated, I have observed an increase of 1245 cc. in total circulating blood in one patient and have calculated that it must have been about this great in another. The value of such increases from the standpoint of hemodynamics is obvious. It is indeed, quite striking that the secretion of the thyroid gland with its marked influence on oxygen consumption should produce such marked changes in the carriers of oxygen and carbon dioxide. As Barcroft (47) has so well put it "The blood volume should be regarded not as aliquot part of the body weight but as a physiological variable which is adjusted to the work required of it."

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nance dose produce proportionately much smaller increases in plasma volume and basal metabolism than the maintenance dose

The plasma volume changes in myxedema are contrasted with those in cardiac edema in which plasma volume increases with increasing edema. Other differences in the two are noted

In particular I wish to thank Dr. James Howard Means for his interest in this work and for help in the preparation of the manuscript. I also wish to thank Dr. Francis W. Peabody for giving me a place to work while I was a house officer

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DIHYDROXYACETONE STUDIES

I ITS RESPIRATORY AND CARBOHYDRATE METABOLISM IN NORMAL MEN¹

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(Received for publication April 28, 1926)

INTRODUCTION

The triose, dihydroxyacetone, has been known for many years, but its experimental study has been restricted due to difficulty in obtaining the chemical in sufficient quantity. Fischer (1) prepared dihydroxyacetone synthetically, the first step being the formation of α acrose by condensation from formaldehyde which by oxidation with bromine was changed into a mixture of glyceric aldehyde and dihydroxyacetone. He further suggested that by treatment with alkalis condensation would take place to hexoses. Another and the commoner method of preparation was by bacterial action upon glycerol.

Theoretical considerations have been advanced by Buchner and Meisenheimer (2), and by Embden, Schmitz, and Wittenberg (3) to the effect that dihydroxyacetone is probably a normal product in the intermediary metabolism of carbohydrate. The former think that it is a product formed early in the fermentation of glucose, while the latter believe that fructose (levulose) is normally split into d-glyceric aldehyde and dihydroxyacetone. More recently Woodyatt (4) has stated that the trioses are probably normal intermediates in carbohydrate metabolism.

¹ The dihydroxyacetone employed in this work was made by Farbwerke v. Meister Lucius and Bruning, Hoechst a. Main, and was supplied gratis by the Mallinckrodt Chemical Works, Ltd. Montreal.

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gastro-intestinal disturbance, and even when taken in large doses (150 grams per day) has shown no evidence of renal irritation. Experiments indicate that it is absorbed slowly by the rectum, and that when given intravenously up to 25 grams it is well tolerated.

In 1924 Isaac and Adler (8) published a communication showing blood sugar curves in normal and diabetic individuals after the administration of 25 to 50 gram doses of dihydroxyacetone by mouth. These were compared with curves obtained in the same patients after equal quantities of glucose. The results were uniform and showed that in normal people there was no rise in the blood sugar after such doses, while in diabetics the rise was of a much less degree than after a similar dose of glucose. This was confirmed by Rabinowitch (11), who also reported one respiratory quotient curve in a mild diabetic after 100 grams of dihydroxyacetone by mouth. A rise of the R Q from the basal level of 0.706 to 0.817 was obtained in two hours.

The work reported in this paper was undertaken to determine the effect of dihydroxyacetone upon the respiratory metabolism of normal individuals. A series of five normal patients were chosen from the general medical wards who had an undisturbed carbohydrate tolerance. Four of the five dihydroxyacetone experiments were controlled by a similar one using the same quantity of glucose.

EXPERIMENTAL METHODS

The respiratory experiments were all conducted under like circumstances with the patient in the basal state during the morning hours. A preliminary basal hour was obtained as a base line, at the end of which the glucose or dihydroxyacetone were given in one dose unless otherwise stated, this being followed by hourly observations for the next three to four hours. The fluid intake amounted to 200 cc per hour. Expired air was collected for ten minute periods in a Douglas bag, during the last ten minutes of the basal hour, and, after the ingestion of the glucose or dihydroxyacetone, during the final ten minutes of the thirty-minute one-hour, two-hour, and three-hour periods. After a preliminary emptying of the bladder at the start of the basal hour, urines were collected hourly and the urinary nitrogen determined in duplicate by the Gunning modification of the original

That dihydroxyacetone is promptly absorbed and under certain circumstances readily converted into glucose is well known. Lusk (5) has shown that all the trioses are quantitatively converted into glucose in a phlorhizinized dog, and Ringer and Frankel (6) confirmed these findings using a nine gram dose of dihydroxyacetone administered subcutaneously. Carbon determinations in their experiments also confirmed its complete conversion into glucose. Embden (3) has further shown its conversion into glucose by liver perfusion. That the trioses, including dihydroxyacetone, are glycogen formers has been shown by Mostkowsk \acute{y} (7). Isaac and Adler (8) stated that in rats and mice after dihydroxyacetone feeding there is more glycogen in the liver than after similar feeding with glucose.

The relation of dihydroxyacetone to lactic acid formation is not clear. Apparently the muscles, muscle juice, and red blood cells can convert dihydroxyacetone into lactic acid (Embden, Baldes, and Schmitz (9), Woodyatt (4)). Isaac and Adler (8) have shown that in cases with a disturbance of liver function after the ingestion of dihydroxyacetone the blood lactic acid rises to a higher level than in individuals with a normal liver function. This might indicate difficulty in forming glycogen and is supported by the fact that with liver disease the blood sugar rises after the ingestion of dihydroxyacetone, a finding which is absent in normal individuals. They have also noted after giving dihydroxyacetone and insulin that the blood lactic acid and its urinary excretion increase much more than after a like dose of glucose and insulin, and that there is an associated fall in the blood sugar. These findings have been obtained in both normal and diabetic individuals. From this they argue that dihydroxyacetone, under the influence of insulin, is not completely changed into glucose but that it is involved in the process of formation of lactic acid by the liver.

That dihydroxyacetone is antiketogenic would appear from the paper of Ringer and Frankel (6) in which phlorhizinized dogs were used. More recently Rabinowitch (10) has reported a case of diabetic ketosis treated with dihydroxyacetone where an antiketogenic action was observed.

The chemical dihydroxyacetone is a white crystalline substance with a slightly tart taste. It dissolves readily in water. Apparently it is absorbed promptly after being taken by mouth, fails to cause any

dihydroxyacetone every fifteen minutes for two hours produced a slightly delayed rise, the maximum respiratory quotient being attained at the end of the first hour. The subsequent drop was of the same character

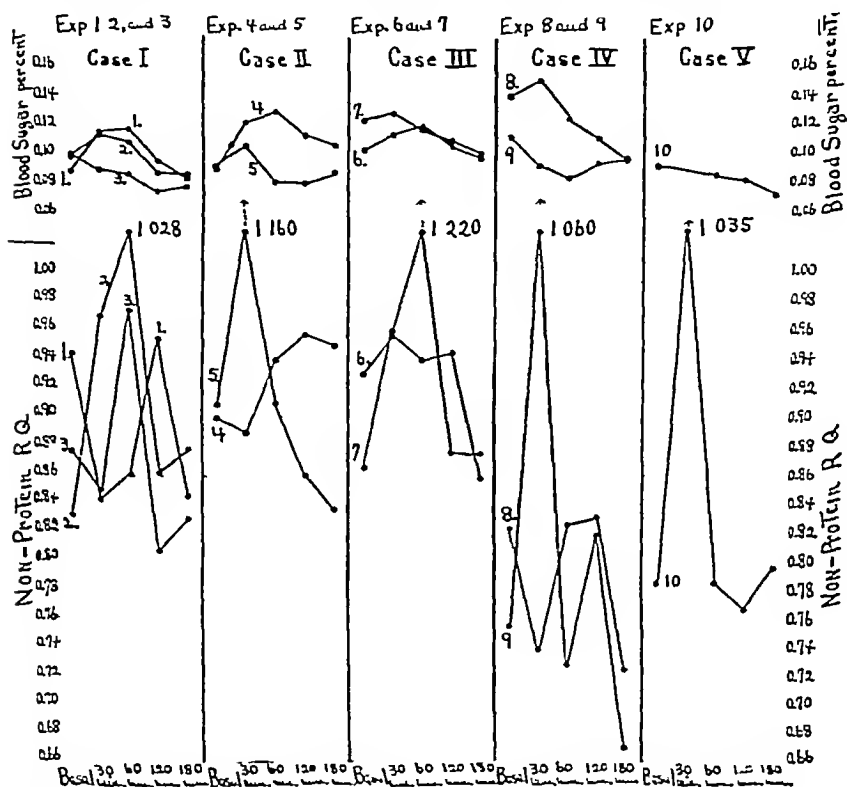


FIG 1 REPRESENTING GRAPHICALLY THE NON-PROTEIN RESPIRATORY QUOTIENT AND BLOOD SUGAR TIME CURVES IN THE EXPERIMENTS RECORDED IN TABLE 1

Experiments 1, 6, and 8 are after 25 grams of glucose, experiment 4 is after 50 grams of glucose. Experiments 2, 7, 9 and 10 are following 25 grams of dihydroxyacetone, experiment 5 is after 50 grams of dihydroxyacetone

with the same increment decrease as in the case of experiment 2, when 25 grams of dihydroxyacetone were given. In four of the cases the observations were controlled by the ingestion of a like dose of glucose,

Kjeldahl technique A blood sample was taken at the end of the basal hour, again in one-half hour, one hour, two hours, etc., and the blood sugar determined by the method of Folin and Wu The expired air was measured in a wet meter and samples were analysed in duplicate by a modified Henderson-Haldane gas analysis apparatus For the qualitative determination of the presence of dihydroxyacetone in the blood and urine the observation that dihydroxyacetone will reduce an alkaline copper solution in the cold was employed In addition a test that Pinoff developed for determining the presence of levulose was used as well It is based on the reduction of ammonium molybdate to an oxide in an acid solution when a blue color is produced (Miller and Taylor (12)) The calculation of the non-protein respiratory quotient was made by subtracting from the total respiratory carbon dioxide and oxygen the protein carbon dioxide and oxygen In the production of one gram of urinary nitrogen 5.91 liters of oxygen are consumed and 4.76 liters of carbon dioxide are given off The non-protein respiratory quotients based on the gas collection of the first thirty minutes of the test period were corrected as for the urinary nitrogen of the complete hour The estimation of the non-protein calories was made according to the tables of Zuntz and Schumberg as modified by Lusk (13) The factor used for the conversion of carbohydrate calories into grams was 3.74, thereby expressing combustion in terms of glucose In the experiments where dihydroxyacetone was present in the urines the amount is expressed in terms of glucose

EXPERIMENTAL FINDINGS

A study of the data obtained in five normal individuals, as summarized in table 1, and depicted graphically in figure 1 shows that there is a material difference in the respiratory and carbohydrate metabolism after like doses of glucose and dihydroxyacetone

The respiratory quotient After the ingestion of 25 or 50 grams of dihydroxyacetone there is a rapid rise in the non-protein respiratory quotient which reaches unity or slightly above in one-half to one hour This rise is followed by a rapid drop to or slightly below the basal level In no case was this rise preceded by a fall in the respiratory quotient In one case, experiment 3, the ingestion of 2 grams of dihy-

Case II Experiment 5 April 23, 1925 Dihydroxyacetone 50 grams									
Basal	10 78	12 12	0 588	0 780	0 910	58 73	9 01	0 092	0
30	14 91	13 61	1 090	—	1 160	67 23	14 94	0 107	0
60	11 28	12 67	0 890	0 424	0 911	61 38	9 30	0 082	0 1
120	10 58	12 47	0 848	0 419	0 860	59 76	6 84	0 082	0
180	9 27	11 13	0 834	0 332	0 837	53 21	5 31	0 089	0
240	9 43	11 09	0 847	0 316	0 860	53 38	6 30	0 089	0
Case III Experiment 6 February 3, 1926 Male Age 18 Surface area 1 625 sq m Glucose 25 grams									
Basal	11 55	12 64	0 914	0 275	0 930	61 98	11 32	0 104	—
30	12 01	12 85	0 933	—	0 957	63 23	13 60	0 114	—
60	11 95	12 98	0 920	0 316	0 940	63 63	11 90	0 120	—
120	12 06	13 08	0 921	0 336	0 945	64 16	12 40	0 106	—
180	10 95	12 82	0 854	0 262	0 858	61 99	7 85	0 098	—
Case III Experiment 7 February 4, 1926 Dihydroxyacetone 25 grams									
Basal	10 21	11 94	0 858	0 282	0 866	57 67	7 54	0 124	0
30	11 42	12 20	0 937	—	0 960	59 93	12 00	0 129	—
60	15 95	13 77	1 160	0 322	1 220	68 33	16 00	0 118	0
120	10 36	11 95	0 867	0 306	0 876	57 91	7 92	0 110	0
180	10 07	11 68	0 862	0 393	0 875	56 21	7 25	0 101	0
Case IV Experiment 8 January 11, 1926 Male Age 32 Surface area 1 67 sq m Glucose 25 grams									
Basal	9 12	11 46	0 822	0 574	0 823	54 20	1 32	0 140	—
30	8 93	11 87	0 753	—	0 738	55 13	1 32	0 152	—
60	10 30	12 58	0 819	0 439	0 823	59 83	5 32	0 125	—
120	9 85	11 95	0 824	0 487	0 830	56 70	5 14	0 111	—
180	8 88	11 93	0 714	0 499	0 724	55 52	0 71	0 096	—
Case IV Experiment 9 January 11, 1926 Dihydroxyacetone 25 grams									
Basal	8 80	11 11	0 771	0 668	0 754	53 10	1 61	0 112	0
30	12 48	12 51	0 997	—	1 060	61 40	12 71	0 093	—
60	11 10	12 48	0 889	0 552	0 727	60 40	9 52	0 084	0
120	9 23	11 32	0 815	0 112	0 818	53 70	4 34	0 094	0
180	7 90	11 19	0 706	0 520	0 669	51 48	0	0 097	0

TABLE 1
Simultaneous respiratory exchange and blood sugar time curves in normals

Time	CO ₂ per hour	O ₂ per hour	Total R.Q.	Nitrogen per hour	Non protein R.Q.	Calories per hour	Carbohydrate per hour	Blood sugar	Dihydroxy acetone in blood	Dihydroxy acetone in urine	Remarks
	liters	liters		grams			grams	per cent		per cent	
Basal	11 23	12 37	0 904	0 551	0 944	59 95	9 95	0 089	—	—	Case I Experiment 1 May 12, 1925 Male Age 15 Surface area 1 43 sq m Glucose 25 grams
30	10 69	12 83	0 833	—	0 843	61 20	6 12	0 116	—	—	
60	11 02	12 98	0 849	0 517	0 861	62 00	7 03	0 118	—	—	
120	11 68	12 40	0 939	0 200	0 955	61 30	13 50	0 096	—	—	
180	10 23	12 29	0 832	0 691	0 846	58 15	5 25	0 084	—	—	
Basal	10 42	12 60	0 827	0 389	0 832	60 10	5 92	0 098	0	0	Case I Experiment 2 May 13, 1925 Dihydroxyacetone 25 grams
30	12 78	13 53	0 945	—	0 970	66 75	13 82	0 115	0	—	
60	12 61	12 73	0 991	0 360	1 028	62 95	14 30	0 109	0	0	
120	10 41	12 17	0 856	0 240	0 862	58 76	7 66	0 088	0	0	
180	10 60	12 23	0 866	0 334	0 879	59 05	8 11	0 087	0	0	
Basal	10 89	12 67	0 859	0 505	0 876	60 72	7 52	0 100	0	0	Case I Experiment 3 May 15, 1925 Dihydroxyacetone 2 grams every 15 minutes for 2 hours
30	10 80	12 89	0 838	—	0 850	61 45	6 32	0 090	0	—	
60	12 63	13 54	0 934	0 560	0 974	66 25	12 60	0 088	0	0	
120	10 28	12 73	0 809	0 594	0 808	60 03	4 29	0 076	0	0	
180	10 43	12 76	0 817	1 115	0 830	59 38	3 49	0 079	0	0	
Basal	9 89	11 28	0 877	0 464	0 900	54 33	7 44	0 091	—	—	Case II Experiment 4 April 21, 1925 Male Age 26 Surface area 1 525 sq m Glucose 50 grams
30	11 22	12 84	0 875	—	0 890	62 09	8 45	0 123	—	—	
60	11 50	12 60	0 913	0 441	0 940	61 42	10 59	0 130	—	—	
120	11 28	12 17	0 926	0 429	0 958	59 51	11 02	0 114	—	—	
180	11 16	11 77	0 948	0 035	0 951	58 59	12 84	0 107	—	—	
240	10 47	11 61	0 900	0 048	0 904	57 08	12 68	0 097	—	—	

experiments 1, 4, 6, and 8. In three of these experiments, 1, 4, and 8 the respiratory quotients showed a decrease during the first one-half hour, as originally noted by Zuntz and Mering (14). This early fall in the respiratory quotient after the administration of glucose is considered to have a relation to the degree of saturation of the glycogen depots. Subsequently, the maximum increment increase was attained at the end of the first or second hour, but in no case was the rise as great as after a like dose of dihydroxyacetone. In the four experiments with glucose the average maximum increment increase

TABLE 2
Normals. Increase in heat production over basal level

Experiment Number	Dose	Increase calories per hour		Time attained
		Glucose	Dihydroxyacetone	
	<i>grams</i>	<i>per cent</i>	<i>per cent</i>	<i>minutes</i>
1	25	3.6	—	60
2	25	—	11.2	30
3	*	—	9.0	60
4	50	14.0	—	30
5	50	—	14.5	30
6	25	3.4	—	120
7	25	—	18.3	60
8	25	10.8	—	60
9	25	—	16.0	30
10	25	—	20.5	30
Average		10.6	16.1	

* Dihydroxyacetone 2 grams every 15 minutes for 2 hours.

in the non-protein respiratory quotient was 0.026, while in the five dihydroxyacetone experiments it was 0.271. The fact that the non-protein respiratory quotients rose above unity after administration of single large doses, 25 to 50 grams of dihydroxyacetone, would suggest change of dihydroxyacetone or its conversion product to fat as shown by Lusk (15) to follow the "metabolism of carbohydrate plethora." The fall of the non-protein respiratory quotient to 0.669 in the third hour in experiment 9 may possibly be explained by the storage of glucose derived from protein.

The total metabolism. The increase in total metabolism is more

Otherwise there would not be such a difference between the parallel experiments

The impossibility of detecting the dihydroxyacetone qualitatively in the blood stream would argue for its prompt conversion or removal. Also, the fact that the blood sugar shows a very slight increment increase, and in some cases a progressive fall, would also indicate its rapid removal or conversion.

The similarity of these results with those as reported by Benedict and Carpenter (16) after levulose is striking. In seven experiments on seven normal subjects using 100 grams of levulose they found an average increase of heat production of 15 per cent as against our 16 per cent in six experiments with dihydroxyacetone (table 2). The total respiratory quotient in their cases had an average maximum increase of 18 points, as compared with a similar increase of 22 points after dihydroxyacetone. Their highest total respiratory quotient was 1.11 and values of 1.07 to 1.09 were of frequent occurrence. Following dihydroxyacetone the maximum total respiratory quotient was 1.16 (experiment 7) and in one other instance (experiment 5) a value of 1.09 was obtained.

CONCLUSIONS

1. In normal men dihydroxyacetone when given in 25 or 50 gram doses causes a more rapid and a greater carbohydrate combustion than does the same dose of glucose.

2. The total metabolism is usually elevated more rapidly and to a higher level of heat production after dihydroxyacetone than after a like dose of glucose.

3. The blood sugar presents less increment increase after dihydroxyacetone than after a like dose of glucose.

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marked and usually occurs more rapidly after dihydroxyacetone than after a like dose of glucose. From table 2 it can be seen that in all instances except experiments 4, and 5, comparable experiments upon the same case gave a greater and earlier rise in heat production with dihydroxyacetone than after the same dose of glucose. In experiments 4 and 5, both after a dose of 50 grams, the increase in heat production was practically the same 14.0 and 14.5 per cent, and in both instances the maximum level was reached in thirty minutes. The average maximum increase of heat production in the case of glucose was 10.6 per cent as against 16.1 per cent after dihydroxyacetone. In experiment 3 when dihydroxyacetone was given at the rate of 2 grams every fifteen minutes for 2 hours the rise in total metabolism was delayed, its peak being reached at the end of sixty minutes, and again with a percentage increase of 9 per cent, appreciably greater than after the single dose of 25 grams of glucose in the same case, experiment 1. In experiment 4 (50 grams glucose) the rise in total metabolism took place coincidentally with a slight fall in the respiratory quotient, subsequently declining as the respiratory quotient rose.

The blood sugar The blood sugar curves obtained synchronously with the respiratory data show a consistently lower level after the dihydroxyacetone than after a like dose of glucose. In two instances experiments 5 and 10, a trace of dihydroxyacetone was detected in the urine of the first test hour, a demonstration of its probable low renal threshold.

DISCUSSION

From the above data it would appear that dihydroxyacetone when ingested by normal men in doses of 25 or 50 grams results in a fundamentally different metabolism from that of glucose. The immediate and marked rise of the respiratory quotient would suggest rapid carbohydrate combustion, or combustion of a conversion product such as lactic acid. That combustion takes place is further supported by the accelerated increase in heat production which parallels the rise in the respiratory quotient.

These studies indicate that at least the greater part of the dihydroxyacetone escapes conversion into glucose in the alkaline duodenum.

DIHYDROXYACETONE STUDIES

II ITS RESPIRATORY AND CARBOHYDRATE METABOLISM IN DIABETES MELLITUS

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(Received for publication April 27, 1926)

In the first paper (1) it was shown that in normal persons dihydroxyacetone given by mouth in 25 or 50 gram doses caused a more rapid and a greater carbohydrate combustion than the same dose of glucose. The elevation of the total metabolism was more marked after the dihydroxyacetone and there was little if any increment increase in the blood sugar.

In this report similar studies have been applied to cases of diabetes mellitus. The methods employed have been the same. There have been compared in the same cases the respiratory exchange and the blood sugar curves after like doses of glucose and dihydroxyacetone. Fifteen experiments upon seven cases of diabetes mellitus are reported. In all cases except one the amount of glucose or dihydroxyacetone ingested was 25 grams. In the one exception it was 50 grams. Comparable experiments after like amounts of glucose and dihydroxyacetone were performed in four instances, cases I, II, III, and IV. In the remaining six experiments dihydroxyacetone alone was given. In case I a three hour control experiment after water alone is included.

THE TYPE OF CASE STUDIED

In order to clarify certain of the experimental data it is necessary to consider the degree of severity of the carbohydrate disturbance in the cases studied. In table 1 are presented the pertinent data on each of the seven cases.

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In table 3 and figure 2, are reported seven experiments upon four cases of diabetes mellitus. In each experiment 25 grams of dihydroxyacetone were given. In cases IV and V comparable experiments were performed at intervals with a progressively increasing carbohydrate intake. These results demonstrate clearly that the increment increase of the respiratory quotient is influenced by the glycogen reserve of the body (table 4). If the glycogen reserves are depleted the ingestion of 25 grams of dihydroxyacetone results in very little combustion, but the added carbohydrate is largely stored. In the same case as the diet was progressively increased in its carbohydrate content the ingestion of the same amount of dihydroxyacetone caused a prompt and marked rise in the respiratory quotient. It would appear from table 4 that the maximum increment increase of the respiratory quotient is attained more rapidly when the past diet has contained more carbohydrate. It is to be expected that this point would only be applicable in those cases of diabetes mellitus who have the ability to store glycogen.

In Experiment 14 a respiratory quotient of unity was attained in 30 minutes. This was a mild adult diabetic who was taking a diet containing a total glucose yield of 128 grams. The slight increment increase in the blood sugar confirms the mildness of the diabetes. Experiment 15 was done upon a young girl, aged 13, with mild diabetes, following a period of low carbohydrate intake. At the time of the test the total glucose yield of the diet was 76 grams.

The average maximum increment increase of the non-protein respiratory quotient in the eleven experiments after dihydroxyacetone was 0.138. The excessively low non-protein respiratory quotients obtained in experiments 5, 7, and 14, namely 0.642, 0.661, and 0.650 respectively, during the second or third hours may be explained by the storage of carbohydrate derived from protein.

The total metabolism

In the comparable experiments after glucose and dihydroxyacetone (table 2) the increase of the total heat production over the basal level was more marked after dihydroxyacetone. There was one exception to this finding, case II, experiments 4 and 5, when the increase was 21.3 and 11.4 per cent after glucose and dihydroxyacetone respectively.

EXPERIMENTAL DATA

The experimental data are tabulated in tables 2 and 3, and in part depicted graphically in figures 1 and 2, respectively. Eight of the comparable experiments after like amounts of glucose or dihydroxyacetone are grouped in table 2 and figure 1. In table 3 and figure 2, there are recorded experiments after dihydroxyacetone alone, cases IV, and V, having multiple observations performed at different periods in the treatment of the case.

TABLE 1
Carbohydrate tolerance upon discharge from hospital

Case number	Age	Diet		Blood sugar fasting	Insulin per day	Severity of case
		'G'*	Total calories			
	years	grams		per cent	units	
I	42	95.8	2205	0.140	24	Moderate
II	55	98.9	2011	0.122	10	Moderate
III	31	112.8	2166	0.142	18	Moderate
IV	17	112.8	2166	0.125	0	Mild
V	13	90.8	1898	0.166	24	Moderate
VI	57	128.6	2248	0.136	0	Mild
VII	13	103.3	1729	0.121	0	Mild

* "g" = glucose yield of diet (Woodyatt)
= $CHO + 0.58P + 0.1F$

The non-protein respiratory quotient

After 25 or 50 grams of dihydroxyacetone there was a greater increment increase in the non-protein respiratory quotient than after the same amount of glucose (table 2 and figure 1). In all four instances after the dihydroxyacetone the maximum increase in the non-protein respiratory quotient was attained at the end of thirty minutes. The rapidity of elevation after the glucose was not consistent, the maximum non-protein respiratory quotient being attained in from thirty to one hundred and twenty minutes. In these four comparable cases the average maximum increment increase in the non-protein respiratory quotient after glucose was 0.048, and after dihydroxyacetone it was 0.126. It should be noted that the time interval between the two experiments on the same case was not more than three days.

In table 3 and figure 2, are reported seven experiments upon four cases of diabetes mellitus. In each experiment 25 grams of dihydroxyacetone were given. In cases IV and V comparable experiments were performed at intervals with a progressively increasing carbohydrate intake. These results demonstrate clearly that the increment increase of the respiratory quotient is influenced by the glycogen reserve of the body (table 4). If the glycogen reserves are depleted the ingestion of 25 grams of dihydroxyacetone results in very little combustion, but the added carbohydrate is largely stored. In the same case as the diet was progressively increased in its carbohydrate content the ingestion of the same amount of dihydroxyacetone caused a prompt and marked rise in the respiratory quotient. It would appear from table 4 that the maximum increment increase of the respiratory quotient is attained more rapidly when the past diet has contained more carbohydrate. It is to be expected that this point would only be applicable in those cases of diabetes mellitus who have the ability to store glycogen.

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The total metabolism

In the comparable experiments after glucose and dihydroxyacetone (table 2) the increase of the total heat production over the basal level was more marked after dihydroxyacetone. There was one exception to this finding, case II, experiments 4 and 5, when the increase was 21.3 and 11.4 per cent after glucose and dihydroxyacetone respectively.

TABLE 2
Simultaneous respiratory exchange and blood sugar time curves in diabetes mellitus

Time	CO ₂ per hour	O ₂ per hour	Total R Q	Nitrogen per hour	Non protein R Q	Calories per hour	Carbohydrate per hour	Blood		Urine		Remarks
								Sugar	Dihydroxy acetone	Glucose	Dihydroxy acetone	
Basal	7 05	9 18	0 768	0 251	0 761	43 26	1 91	0 182	—	0	—	Case I Experiment 1 May 27, 1925 Male Age 42 Surface area 1 14 sq m Control 200 cc water every hour
30	7 10	9 27	0 764	—	0 762	43 86	2 15	—	—	—	—	
60	6 70	9 01	0 743	0 133	0 738	42 34	1 17	0 184	—	0	—	
120	6 76	8 82	0 766	0 258	0 760	41 46	1 78	0 176	—	0	—	
180	6 77	8 82	0 767	0 250	0 761	41 53	1 82	0 170	—	0	—	
Basal	6 68	9 21	0 724	0 171	0 716	42 95	0 35	0 130	—	0	—	Case I Experiment 2 April 24, 1925 Glucose 50 grams Surface area 1 45 sq m
30	7 85	10 69	0 733	—	0 715	49 74	0 34	0 210	—	—	—	
60	7 83	10 47	0 746	0 364	0 730	48 94	0 86	0 280	—	0 98	—	
120	7 56	9 69	0 781	0 272	0 775	45 79	2 43	0 285	—	2 40	—	
180	7 51	9 62	0 781	0 513	0 770	44 79	1 79	0 242	—	3 03	—	
240	7 24	9 25	0 781	0 081	0 780	44 07	2 86	0 198	—	1 18	—	
Basal	6 70	9 21	0 725	0 391	0 701	42 68	0	0 117	0	0	0	Case I Experiment 3 April 27, 1925 Dihydroxy acetone 50 grams Surface area 1 45 sq m
30	9 40	11 31	0 830	—	0 838	53 93	5 20	0 226	Trace	—	—	
60	9 02	11 36	0 792	0 402	0 790	53 68	3 29	0 218	0	0 33	Trace	
120	8 03	10 89	0 736	0 170	0 730	51 11	1 02	0 188	0	0 15	0	
180	6 96	9 68	0 720	0 112	0 711	45 28	0 19	0 164	0	0 22	0	
240	6 54	9 49	0 688	0 346	0 656	43 98	0	0 146	0	0 18	0	
Basal	7 98	11 23	0 710	0 500	0 677	51 67	0	0 246	—	0	—	Case II Experiment 4 June 9, 1925 Male Age 55 Surface area 1 63 sq m Glucose 25 grams
30	9 89	13 53	0 730	—	0 704	62 57	0	0 264	—	—	—	
60	9 04	12 14	0 743	0 610	0 720	56 27	0 51	0 339	—	0 49	—	
120	9 09	12 36	0 735	0 815	0 692	56 80	0	0 303	—	1 03	—	
180	9 40	13 27	0 707	0 636	0 671	60 87	0	0 261	—	0 69	—	
240	—	—	—	—	—	—	—	0 254	—	0 49	—	

Case II Experiment 5 June 11, 1925 Surface area 1 625 sq m Dihydroxyacetone 25 grams									
Basal	9 95	13 60	0 732	0 376	0 719	63 46	0 63	0 141	0
30	11 98	14 81	0 809	—	0 810	70 71	6 00	0 188	—
60	9 00	12 80	0 702	0 370	0 681	59 41	0	0 168	0
120	8 02	11 80	0 681	0 461	0 642	54 03	0	0 142	0
180	8 27	11 92	0 693	0 466	0 660	54 75	0	0 123	0
240	—	—	—	—	—	—	—	0 133	—
Case III Experiment 6 July 2, 1925 Male Alb 31 Surface area 1 605 sq m Glucose 25 grams									
Basal	8 00	11 29	0 708	0 237	0 696	52 58	0	0 190	0
30	8 37	11 32	0 739	—	0 725	52 76	0 77	0 279	—
60	8 33	11 41	0 730	0 354	0 713	53 16	0 26	0 267	1 32
120	8 26	11 52	0 717	0 437	0 692	53 30	0	0 246	2 12
180	7 51	10 65	0 705	0 326	0 683	49 25	0	0 220	0 41
Case III Experiment 7 June 30, 1925 Dihydroxyacetone 25 grams									
Basal	7 98	10 25	0 777	0 295	0 772	48 43	2 55	0 226	0 29
30	10 39	12 02	0 822	—	0 830	59 76	5 23	0 278	—
60	8 76	12 07	0 726	0 569	0 698	55 73	0	0 306	2 68
120	8 02	11 27	0 712	0 599	0 669	51 68	0	0 252	2 76
180	7 97	11 48	0 691	0 450	0 661	52 73	0	0 231	1 71
Case IV Experiment 8 January 7, 1926 Male Alb 17 Surface area 1 515 sq m Glucose 25 grams									
Basal	7 76	10 37	0 750	0 279	0 738	48 60	1 24	0 125	0
30	7 47	10 60	0 712	—	0 731	49 69	1 03	0 156	—
60	8 39	10 61	0 790	0 252	0 766	50 29	3 32	0 192	—
120	8 51	10 66	0 799	0 235	0 795	50 53	3 76	0 174	—
180	8 02	10 86	0 739	0 235	0 729	50 83	0 96	0 166	0
Case IV Experiment 9 January 5, 1926 Dihydroxyacetone 25 grams									
Basal	7 64	10 53	0 725	0 450	0 696	48 75	0	0 105	0
30	10 60	11 71	0 901	—	0 915	57 34	9 78	0 131	—
60	8 31	10 99	0 759	0 258	0 750	51 61	1 87	0 123	0
120	8 14	10 95	0 743	0 316	0 730	51 18	0 96	0 104	0
180	7 73	10 38	0 750	0 228	0 735	48 65	1 16	0 079	0

TABLE 3
Simultaneous respiratory exchange and blood sugar time curves in diabetes mellitus

Time minutes	CO ₂ per hour	O ₂ per hour	Total R Q	Nitro- gen per hour	Non protein R Q	Calo- ries per hour	Carbo- hydrate per hour	Blood		Urine		Remarks
								Sugar	Dihy- droxy acetone	Glucose	Dihy- droxy- acetone	
Basal	8 73	11 50	0 759	0 616	0 747	53 50	1 44	0 166	0	0	0	Case IV Experiment 10 December 15, 1925 Male Age 17 Surface area 1 515 sq m Dihydroxyacetone 25 grams
30	10 42	13 90	0 751	—	0 737	65 05	1 58	—	—	—	—	
60	9 34	11 71	0 797	0 417	0 795	55 35	3 74	0 260	0	0 47	Trace	
120	8 12	11 38	0 714	0 308	0 696	52 87	0	0 242	0	0	0	
180	7 69	10 79	0 713	0 288	0 696	50 14	0	0 236	0	0	0	
Basal	8 49	11 01	0 769	0 456	0 759	51 56	1 98	0 092	0	0	0	Case IV Experiment 11 December 29, 1925 Dihydroxyacetone 25 grams
30	8 02	11 00	0 730	—	0 708	50 92	0	0 107	0	—	—	
60	10 01	11 73	0 854	0 409	0 865	56 32	6 78	0 144	0	0	0	
120	8 33	11 58	0 721	0 386	0 699	53 72	0	0 134	0	0	0	
180	7 86	10 97	0 716	0 269	0 701	51 03	0	0 094	0	0	0	
Basal	7 64	10 53	0 725	0 450	0 696	48 75	0	0 105	0	0	0	Case IV Experiment 9 January 5, 1926 Dihydroxyacetone 25 grams
30	10 60	11 74	0 901	—	0 915	57 34	9 78	0 131	0	—	—	
60	8 34	10 99	0 759	0 258	0 750	51 64	1 87	0 123	0	0	0	
120	8 14	10 95	0 743	0 316	0 730	51 18	0 96	0 104	0	0	0	
180	7 73	10 38	0 750	0 228	0 735	48 65	1 16	0 079	0	0	0	
Basal	6 84	8 08	0 846	0 134	0 851	38 95	4 96	0 216	0	0	0	Case V Experiment 12 December 2 1925 Male Age 13 Surface area 1 19 sq m Dihydroxyacetone 25 grams
30	8 20	9 66	0 848	—	0 851	46 69	6 00	0 244	0	—	—	
60	8 66	10 12	0 855	0 109	0 858	49 09	6 60	0 252	0	0	0	
120	7 04	9 64	0 730	0 117	0 725	45 19	0 74	0 232	0	0	0	
180	7 06	9 57	0 738	0 176	0 730	44 88	0 90	0 208	0	0	0	

Case V Experiment 13 January 6, 1926 Dihydroxyacetone 25 grams											
Basal	7 42	9 51	0 780	0 414	0 776	14 71	2 24	0 117	0	0	0
30	10 83	11 72	0 925	—	0 910	57 54	11 14	0 238	0	—	—
60	9 36	11 53	0 810	0 224	0 811	55 19	4 90	0 226	0	0 13	+
120	7 72	10 36	0 743	0 404	0 725	48 30	0 66	0 188	0	0	0
180	7 66	10 32	0 741	0 325	0 729	46 22	0 65	0 135	0	0	0
Case VI Experiment 14 June 8, 1925 Male Alge 57 Surface area 1 68 sq m Dihydroxyacetone 25 grams											
Basal	5 99	8 58	0 698	0 206	0 674	44 20	0	0 147	0	0	0
30	9 89	10 07	0 985	—	1 029	49 85	10 80	0 150	0	—	—
60	8 27	10 09	0 817	0 356	0 825	47 94	4 33	0 151	0	0	0
120	7 05	9 73	0 724	0 221	0 710	44 34	0 11	0 150	0	0	0
180	6 70	10 04	0 667	0 204	0 650	46 16	0	0 130	0	0	0
Case VII Experiment 15 November 27, 1925 Female Alge 13 Surface area 1 105 sq m Dihydroxyacetone 25 grams											
Basal	6 24	7 82	0 798	0 335	0 795	36 87	2 37	0 077	0	0	0
30	7 97	8 94	0 891	—	0 905	43 61	7 05	0 119	0	—	—
60	7 55	8 79	0 858	0 206	0 867	42 46	5 93	0 130	0	0	0
120	6 68	8 12	0 831	0 192	0 825	38 78	3 79	0 111	0	0	0
180	6 15	8 07	0 799	0 242	0 787	38 64	2 46	0 075	0	0	0

The basal heat production in Experiment 4 was open to some question. However, without excluding this finding the average rise in heat production after glucose was 11.2 per cent, and after dihydroxyacetone 20.0 per cent. In the eleven experiments with dihydroxy

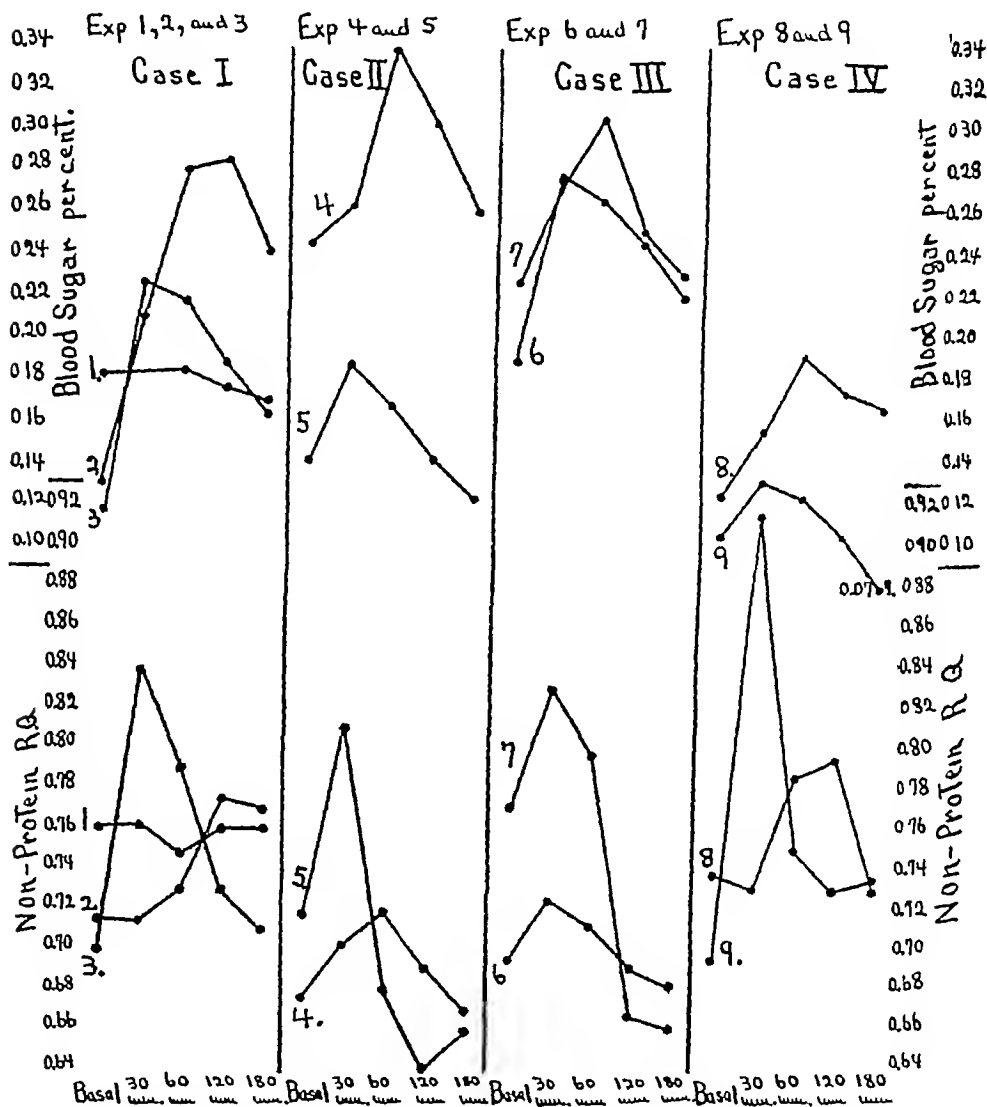


FIGURE 1 REPRESENTING GRAPHICALLY THE NON-PROTEIN RESPIRATORY QUOTIENT AND BLOOD SUGAR TIME CURVES IN THE EXPERIMENTS RECORDED IN TABLE 2

Experiment 1 is a control, 200 cc. of water only being given each hour. Experiment 2 is after 50 grams of glucose. Experiments 4, 6, and 8 are after 25 grams of glucose. Experiment 3 is after 50 grams of dihydroxyacetone. Experiments 5, 7, and 9 are after 25 grams of dihydroxyacetone.

acetone the average increase in heat production over the basal level was 19.4 per cent. In all except two of these experiments the maximum increase in heat production was attained in thirty minutes,

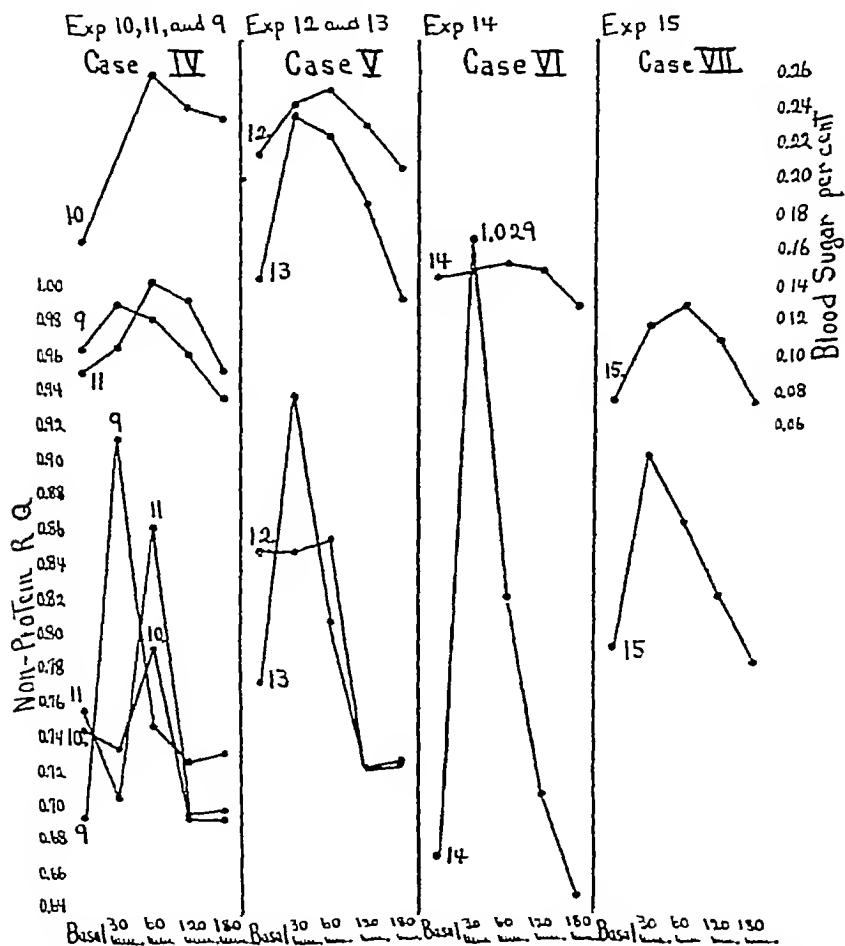


FIGURE 2 REPRESENTING GRAPHICALLY THE NON-PROTEIN RESPIRATORY QUOTIENT AND BLOOD SUGAR TIME CURVES IN THE EXPERIMENTS RECORDED IN TABLE 3

Each experiment is after 25 grams of dihydroxyacetone

while in the two (experiments 11 and 12), it was reached in sixty minutes

In the four glucose experiments the maximum total metabolism was reached in thirty minutes in experiments 2 and 4, in two hours in experiment 6, and in three hours in experiment 8. It should be noted that in cases IV and V in which multiple experiments were done with 25 grams of dihydroxyacetone that the rise in heat production did not parallel the rise in the respiratory quotient. This is shown in table 5.

TABLE 4

Influence of carbohydrate content of diet upon increment increase of respiratory quotient

	Experiment number	'G' of diet	Non protein R Q		Date
			Maximum increment increase	Time attained	
		grams		minutes	
IV	10	45.7	0.048	60	December 15, 1925
	11	96.8	0.106	60	December 29, 1925
	9	112.8	0.219	30	January 5, 1926
V	12	69.0	0.007	60	December 2, 1925
	13	91.0	0.164	30	January 6, 1926

"g" of diet = $C H O + 0.58 P + 0.17$ (Wood, att)

TABLE 5

Relation between increase of non-protein R Q and total metabolism

Case number	Experiment number	Maximum increase	
		Non protein R.Q.	Total calories
			per cent
IV	10	0.048	21.8
	11	0.106	9.0
	9	0.219	17.5
V	12	0.007	26.0
	13	0.164	28.5

The blood sugar

After 25 or 50 grams of dihydroxyacetone the increment increase of the blood sugar was consistently less than after the same dose of glucose (table 2 and chart 1). In most of the cases the difference was marked.

Carbohydrate combustion rate

Inspection of tables 2 and 3 will show the greatly accelerated carbohydrate combustion rate which followed the administration of dihydroxyacetone. In the comparable experiments (table 2) in each instance it greatly exceeded that in the control experiment with glucose. In experiments 9, 13, and 14, and 15 it temporarily reached practically a normal carbohydrate utilization rate, namely 9.78, 11.14, 10.80, and 7.05 grams per hour respectively.

Dihydroxyacetone in blood and urine

Dihydroxyacetone determined qualitatively was detected in the blood only in experiment 3. This was in the thirty-minute specimen. In three experiments, nos. 3, 10, and 13 it was found in the urine in a small quantity in the one hour specimen.

SUMMARY AND CONCLUSIONS

A study of fourteen experiments on seven cases of mild and moderately severe diabetes mellitus would indicate that the respiratory and carbohydrate metabolism of dihydroxyacetone is fundamentally different from that of glucose. For a proper evaluation of the changes in the respiratory quotient after dihydroxyacetone ingestion the carbohydrate content of the previous diet must be known.

The average maximum increment increase of the non-protein respiratory quotient after glucose (4 experiments) was 0.048, while after dihydroxyacetone (10 experiments) it was 0.130.

The average increase of total metabolism after glucose (4 experiments) was 11.2 per cent, while after dihydroxyacetone (10 experiments) it was 19.6 per cent.

The blood sugar showed a smaller increment increase after dihydroxyacetone than after glucose.

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THE EFFECT OF ORALLY ADMINISTERED HYDROCHLORIC ACID UPON THE GASTRIC CONTENTS IN NORMAL INDIVIDUALS AND IN PATIENTS WITH ACHLORHYDRIA*

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(Received for publication May 3, 1926)

The adequacy of the customary dose of dilute hydrochloric acid given to patients with gastric anacidity has been questioned by a number of observers. In 1907, Leo (1) found that a dose of hydrochloric acid corresponding to 2.5 cc (U. S. P. dilute) was the smallest that was ever followed by free hydrochloric acid in the gastric contents, while greater doses, up to a maximum of 12 cc., at times failed to produce free acid. In 1918, Crohn (2) observed that small single doses of hydrochloric acid given to achlorhydria patients were promptly neutralized, and only after the administration of larger doses (1.6 to 2.6 cc.) was free hydrochloric acid found to be present for a time in the fractionally removed gastric contents. For many years the constant finding of gastric anacidity in cases of pernicious anemia has led to speculation as to the possible etiologic relation of achlorhydria to this disease. Hurst (3), who favors such a relationship, has recently emphasized the importance of overcoming the anacidity by giving 4 to 6 cc. of dilute hydrochloric acid as a routine dose. His co-worker, Shaw (4) in a study of five cases of pernicious anemia, found, by fractional gastric analysis, little or no free hydrochloric acid after the administration of 4 cc. of dilute hydrochloric acid, while 8 cc. usually gave sufficient free acid to have a germicidal effect. Vanderhoof (5), in a clinical study of seven cases of combined sclerosis with achlorhydria but without anemia, reported marked clinical

*Read in abstract before the Association of American Physicians at Washington, D. C., May, 1925.

improvement after the continued administration of 4 to 8 cc of dilute hydrochloric acid, and emphasized the importance of adequate dosage of acid

It, therefore, occurred to us to study in detail the effects of various doses of hydrochloric acid upon the gastric contents of individuals with and without achlorhydria, and to attempt to determine the fate of the acid given

In a study of this problem, a number of factors must be taken into consideration. In the first place, it has been amply shown (Bennett and Ryle (6), Talbot (7), Rehfuess et al (8)) that wide variations in both free and total acidity may occur in supposedly normal individuals. The mechanism by which the gastric acid is neutralized is a complex one, and by no means completely understood, involving as it does in addition to neutralization by swallowed saliva and mucus and possibly dilution by gastric secretion, also the regurgitation of alkaline duodenal contents, and the secretion of an alkaline fluid by the pars pylorica, as suggested by Ivy and Oyama (9). In order to determine the part played by some of these factors, we studied in addition to total and free acid in the gastric contents, the hydrogen-ion concentration, the total-chloride concentration, peptic activity, and in some instances the tryptic activity.

It must be stated at the outset that the various values obtained in gastric contents removed by the fractional method are not advanced as representative of a uniform composition of the stomach contents of that part of the stomach (the antrum) from which we tried as a rule to remove our samples. We do believe, however, that the variations of these values, and the curves derived therefrom, can give some information as to the normal and pathological physiology of the stomach, and the influence on these processes of a therapeutic agent such as hydrochloric acid.

MATERIAL

Studies were made on twenty-one patients. Of these, seven had no gastrointestinal symptoms, and have been grouped as "normal cases," six of them showed normal gastric findings, and one was found to have hyperacidity. There was also included in this group one case of hypoacidity, a patient with possible alcoholic gastritis. The

remaining thirteen patients had gastric anacidity, due in nine instances to primary pernicious anemia, in one to a gastric ulcer, proven at operation and possibly syphilitic in origin, in another associated with suspected syphilis of the stomach, while in two patients there was an idiopathic anacidity. One of the latter had a diaphragmatic hernia, while the other suffered from colitis. In nineteen individuals, a fractional gastric analysis was done, with determinations of free and total acidity, pH, and in selected samples, chloride concentration and peptic and tryptic activity. In fourteen patients (four "normal" and ten anacidity cases), such observations were also made after the administration, with the test meal, of dilute hydrochloric acid, U S P, in amounts varying from 1 to 14 cc.

METHODS

The test meal used consisted of two slices of wheat bread with two glasses (about 360 cc) of water. The acid, when given, was usually swallowed by the subject, but in a few instances the mixture of acid and water was introduced into the stomach through the tube, the bread being swallowed first. Specimens of gastric content were withdrawn at varying time intervals, as shown in the tables below. An attempt was made in every instance to pass the tube into the antrum of the stomach.

Most of the samples removed, up to one hour after the meal contained undigested bread, and many were also mixed with varying amounts of mucus and saliva. Accordingly, in every instance, except when the samples were unmixed with bread particles, mucus or saliva, they were centrifuged, and the supernatant liquid used for all determinations.

One or 2 cc of the sample were titrated with 0.02 N NaOH, dimethyl-amidoazobenzol being used as the indicator for free hydrochloric acid, with the permanent yellow taken as end-point, and an alcoholic solution of phenolphthalein as the indicator for total acidity, with the first permanent pink taken as the end-point. The free and total acidity were expressed, as usual, in terms of cubic centimeters of 0.1 N acid per 100 cc of gastric content.

An approximate indication of the pH was obtained by colorimetric comparison with standard buffer solutions, using appropriate indi-

cators The following solutions were made (Clark (10)), pH 1.5 and 2.0 (KCl-HCl mixtures), pH 2.5, 3.0, and 3.5 (phthalate-HCl mixtures), pH 4.0, 4.5, 5.0 and 5.5 (sodium acetate-acetic acid mixtures), pH 6.0, 6.5, 7.0 and 7.5 (phosphate mixtures) The indicators used were thymol blue (pH 1.0 to 2.5), methyl orange (pH 3.0 to 5.0), brom-cresol-purple (pH 5.0 to 7.0), and phenol red (pH 7.0 +)

One drop of the indicator was added to 0.8 cc of the sample of gastric content and to 0.8 cc each of two or more standard solutions with which the sample was to be compared, each portion of standard solution and the gastric content being placed on a porcelain spot-plate The standard solution showing the color nearest to that of the gastric sample was taken as the colorimetric pH of the stomach content A reading accurate to 0.5 pH was considered adequate It is recognized that this colorimetric pH may differ from the true pH of the gastric contents by at least 1.0 because of protein and salt error The variations in pH in which we are interested are, however, so gross that this crude index of gastric pH has some value

Determinations of the chloride content were made on the supernatant fluid of the centrifuged gastric samples by the Volhard method The results are expressed in terms of milliequivalents per liter, as are the results of titration for free hydrochloric acid and total acidity

Pepsin and trypsin determinations were made by the method of Itakura (11) which is described as a modification of the Fuld-Gross method, and is done as follows

Pepsin A casein stock solution is made by dissolving 0.15 gram of dry casein in 50 cc of 0.1 N NaOH, heating, while stirring, on a waterbath to 80°C, placing in a vessel of cold water, and then bringing to 100 cc volume with distilled water 10 cc of the above solution are pipetted into a 100 cc volumetric flask containing 30 cc of 0.1 N HCl, brought to the mark with distilled water, and shaken The resulting solution is an almost clear liquid which may be preserved by the addition of a small amount of chloroform

An acetate mixture is used to precipitate the undigested casein, stop enzyme action, and prevent the precipitation of bile acids It is prepared by titrating 100 cc of 15 per cent NaOH against litmus paper with 6 per cent acetic acid After neutralization, nine-tenths the volume of acid used in the titration is added

To obtain an approximate idea of the degree of peptic activity in the sample of stomach contents, 0.1, 0.01, 0.001, and 0.0001 cc of the sample obtained by serial dilution with 0.01 N HCl are placed in five tubes (preferably 1 cm in diameter

and 10 cm in length), and each is brought to 1 cc by the addition of 0.01 N HCl. Two cubic centimeters of the casein solution are added to each tube which makes the final concentration of casein 0.01 per cent the tubes are stoppered and placed in a water bath at 38°C for a convenient time (This is usually from one-half to one hour)

The digestion of the casein occurs in a medium the pH of which is approximately 1.7

At the end of the half hour or hour period the tubes are removed from water bath, five or six drops of the acetate solution are added to each tube, and they are cooled quickly to room temperature in cold water. The undigested casein will be precipitated in a few minutes, making the solution opaque or hazy, while clarity of the solution is taken as an indication of complete casein digestion. Incubation with the above dilutions of gastric juice gives an idea of the lowest concentration at which digestion is completed, in the time allowed. For a more accurate reading a second series of dilutions of gastric juice is now made—between the dilutions which in the first incubation showed a change from cloudiness to clearness.

The test permits the selection of that critical tube which contains the lowest concentration of gastric juice sufficient to digest completely the standard casein solution in a given time interval and this critical tube is taken as representing a given concentration of pepsin. Itakura defines a unit concentration of pepsin as that concentration which will digest the standard 0.01 per cent casein solution at 38°C and under the condition of the test in one hour. He found that the concentration of pepsin required was inversely proportional to the time allowed for digestion. He expressed the pepsin concentration in the original gastric juice or unknown solution as log *P* defined as follows

$$\text{Log } P = \log \frac{3}{ET}$$

where *E* = cubic centimeters of gastric juice (or unknown solution) contained in the critical tube of the test (total content 3 cc) and *T* = time, in hours, of incubation at 38°C

It must be remembered that log *P*, the expression for peptic activity, being logarithmic, a variation in log *P* of 1.0 represents a ten fold variation in the measured peptic concentration.

For determination of trypsin the method is the same, except that the casein solution is made as follows

To 10 cc of the casein stock solution in a 100 cc volumetric flask are added 50 cc of distilled water and a drop of 0.5 per cent phenolphthalein solution (0.5 gram in 100 cc of 43 per cent alcohol) and this is titrated carefully with 0.1 N hydrochloric acid until the rose-red color disappears. The mixture is then made up to the mark with distilled water. In preparing the tubes for incubation the dilution of gastric content is made up to 1 cc by diluting with distilled water after

neutralization of the gastric content. The final mixture should have a pH of about 7.0. In testing for trypsin it is therefore necessary to neutralize acid present by adding the proper amount of sodium hydroxide. The method is similar in all other respects to the one used for pepsin determination.

For details of the methods, and the derivation of the equation used, the reader is referred to Itakura's article.

CONTROL OBSERVATIONS WITHOUT ACID ADMINISTERED

Normal individuals

Studies were first made in a series of so-called normal individuals. The examinations included a fractional gastric analysis, with determinations of total acidity, free hydrochloric acid, pH and, in selected instances, chloride content and peptic activity. In four of the eight cases so studied, the fasting content showed no free hydrochloric acid, while in the other four, free hydrochloric acid was present in the fasting content. The results in these two groups are tabulated separately.

No free HCl in fasting contents

Table 1 gives the figures obtained in the four cases showing no free hydrochloric acid in the fasting content.

Analysis of table 1 shows

1. The free hydrochloric acid and total acidity curves conform to average normal values, reaching a maximum at between 45 and 90 minutes.

2. pH decreased progressively up to 60 to 75 minutes in all four cases, pH variations roughly paralleled those of the free hydrochloric acid.

3. Chloride content, in the two cases studied, showed a progressive increase, reaching maximal values at 90 minutes, the time of the last determination.

4. Peptic activity, in the one case studied, was within normal limits.

With free HCl in fasting contents

Table 2 gives the findings of similar studies in the subjects showing free hydrochloric acid in the fasting content.

Analysis of table 2 shows

TABLE 1
Control observations in normals without acid administered No free hydrochloric acid in fasting contents

	Fasting content	15 minutes	30 minutes	45 minutes	60 minutes	75 minutes	90 minutes	105 minutes
Experiment 1 Case 1 (Mr R) Chronic arthritis	Total acidity							
	Free HCl	8	qns	62	67	63	49	47
	pH	0	qns	50	40	45	32	33
	Chloride	6.5	qns	2+	2.0	<2.0	2.0	<2.0
Experiment 2 Case 2 (Mr Mel) Chronic arthritis	Total acidity							
	Free HCl	10	37	54	59	48	67	55
	pH	0	12	28	35	20	12	26
	Chloride	6.5	3.0	2.5	2.5	2.5	1.5	<2.0
Experiment 3 Case 3 (Mr W) Colitis (?)	Total acidity							
	Free HCl	8	20	qns	41	16	qns	15
	pH	0	0	qns	16	13	qns	5
	Chloride	7.0	4.6	qns	4.0	4.0	qns	4.0
Experiment 4 Case 4 (Mr P) Alcoholic gastritis (?)	Total acidity							
	Free HCl	6	12	26	45	16	18	12
	pH	0	0	13	16	0	0	0
	Chloride	7.0	6.0	1.0	3.0	1.5	5.0	6.0

* pH = peptic activity. Total acidity, free HCl and chloride are expressed in cubic centimeters of N/10 per 100 cc. which is the same as milliequivalents per liter.

TABLE 2
Control observations in normals without acid administered With free hydrochloric acid in gastric contents

	Fasting content	15 minutes	30 minutes	45 minutes	60 minutes	75 minutes	90 minutes	105 minutes	120 minutes	150 minutes
Experiment 5 Case 5 (Mr P) Sciatica	Total acidity	91	82	72	102	60	q n s	50	—	—
	Free HCl	58	46	32	60	35	q n s	15	—	—
	pH	<2 0	1 5	2 0	<2 0	<2 0	—	—	—	—
	pP	4	3 6	3 6	—	—	—	—	—	—
Experiment 6 Case 6 (Mr B) Chronic arthritis	Total acidity	45	52	36	58	69	39	—	50	43
	Free HCl	15	32	18	36	53	22	—	32	23
	pH	3 0	<2 0	2 0	—	<2 0	2 0	—	2 5	2 5
	Chlonde	118	79	79	—	107 9	—	—	—	98 9
Experiment 7 Case 7 (Mrs W) Neurosis	Total acidity	25	69	110	119	104	106	109	—	—
	Free HCl	14	42	80	95	80	90	94	—	—
	pH	2 0	<2 0	<2 0	<2 0	<2 0	<2 0	<2 0	—	—
	Chlonde	39 1	84 9	3 7	134 5	—	—	135 1	—	—
Experiment 8 Case 8 (Mrs K) Viscerptosis	Total acidity	19	25	47	67	79	89	86	—	—
	Free HCl	11	18	24	40	47	57	62	—	—
	pH	q n s	2 0	<2 0	3 0	<2 0	2 5	<2 0	—	—
	Chlonde	—	36 6	—	—	—	102	—	—	—

1 Both free hydrochloric acid and total acidity curves in the cases showing free hydrochloric acid in the fasting content tend to rise higher than those with no free hydrochloric acid in the fasting content

2 pH was low throughout, falling in the first thirty minutes, and usually rising slightly thereafter pH variations again roughly paralleled those of the free hydrochloric acid

3 In two cases the chloride concentration increased progressively and in a third case was high throughout

4 Peptic activity, in the two cases studied, was greater than in the case in the previous group, it did not, in the one case in which it was tested at intervals increase parallel with the acidity, but, in fact, decreased slightly

Patients with achlorhydria

Similar studies were then made thirteen times in twelve patients with achlorhydria Eight were cases of primary pernicious anemia, two probably syphilis of the stomach, while in two there was no obvious cause for the achlorhydria The figures are given in table 3

Analysis of table 3 shows

1 The total acidity, except in case 17, did not exceed 25 milliequivalents, the variation in total acid was not consistent, but tended to fall slightly in most instances

2 No free hydrochloric acid was present in any case

3 pH remained fairly constant in all except case 17 it was 5.5 or above

4 Chloride concentration in the fasting content was within the range observed in normal individuals but was considerably less than in the normal during the subsequent periods In all instances there was a primary fall in chloride concentration, probably due to dilution of the gastric content by the entrance of the test meal, followed by a rise of varying degree

5 Peptic activity was frequently diminished to 1.5 and when studied at intervals showed practically no variation In one instance, case 17, a patient with syphilis of the stomach, the peptic activity was normal It is interesting to note that in this case alone were there high values for total acidity, and a pH close to 3.0

6 Trypsin was present in all five cases in which a test for it was

TABLE 3
Control observations in cases of anacidity No acid given

	Fasting content	5 minutes	10 minutes	15 minutes	20 minutes	30 minutes	45 minutes	60 minutes	75 minutes	90 minutes	105 minutes
Experiment 9 Case 9 (Mrs B) Primary pernicious anaemia	7 0 6.5	— — —	— — —	17 — 6.5	— — —	11 0 6.5	16 0 6.0	13 0 6.0	10 0 6.0	13 0 6.0	17 0 6.5
Experiment 10 Case 10 (Mr R) Primary pernicious anaemia	25 0 6.5	— — —	— — —	9 0 6.5	— — —	— 0 6.5	6 0 6.5	7 0 6.5	9 0 6.0	8 0 6.5	8 0 6.5
Comp 16*											
Experiment 11 Case 11 (Mrs H) Primary pernicious anaemia	8 0 7.0	— — —	— — —	11 0 7.0	— — —	11 0 6.5	12 0 6.5	13 0 6.5	15 0 —	16 0 6.5	9 0 6.5
Experiment 12 Case 12 (Mr McC) Primary pernicious anaemia	7 0 6.5 42.6 3	— — — — —	— — — — —	7 0 6.5 34.3	— — — —	5 0 6.5 — 2.4	6 0 6.5 —	12 0 7.0 68.8	10 0 6.5 —	12 0 7.0 74.2	11 0 6.5 — 2.4
Experiment 13 Case 13 (Mr C) Primary pernicious anaemia	15 0 — 56.8 3	— — — — —	— — — — —	5 0 6.0 40.1 1.8	— — — — —	qns qns qns	8 0 6.5	12 0 6.0	12 0 6.0	16 0 qns	14 0 qns

Experiment 14 (Case 14) (Mrs M) Primary pernicious anemia	Total acidity Free HCl pH Chloride	5 0 7.5 —	— — — —	— — — —	6 0 6.5 8.7	— — — —	6 0 7.0 —	5 0 7.0 15.2	10 0 6.5 —	5 0 7.5 4.1	qns qns qns qns	
Experiment 15 (Case 15) (Mr II) Primary pernicious anemia	Total acidity Free HCl pH	9 0 (bile) 7.5+	— — —	— — —	15 0 6.5	— — —	5 0 6.5	5 0 6.5	8 0 6.5	11 0 6.5	10 0 6.0	— — —
Experiment 16 (Case 16) (Mrs I) Primary pernicious anemia	Total acidity Free HCl	8 0	— —	— —	18 0	— —	16 0	qns 0	qns 0	8 0	8 0	12 0
Experiment 17 (Case 17) (Miss G) Gastric syphilis	Total acidity Free HCl pH Chloride pH	60 0 3.0 63.0 3.2P	— — — —	— — — —	61 0 3.0 62.1	— — — —	30 0 3.0 —	28 0 3.5 —	26 0 3.5 36.9	26 0 3.5 —	30 0 2.5 —	50 0 3.5 52.7
Experiment 18 (Case 18) (Mr I) Gastric ulcer (syphilitic?)	Total acidity Free HCl pH Chloride pH pH	10 0 7.0 46.4 1.5 0.9	6 0 7.5 17.3 1.5	7 0 7.0 20	6 0 6.5 23.2	7 0 6.5 22.6 1.5 0.9	— — — —	— — — —	— — — —	— — — —	— — — —	— — — —
Experiment 19 (Case 19) (Mr I) Diaphragmatic hernia	Total acidity Free HCl pH Chloride pH	16 0 7.0 71.6 —	— — — —	— — — —	11 0 6.5 70.7 2.8	— — — —	9 0 6.5 —	8 0 7.5 52	12 0 7.0 58.3	9 0 6.0 —	7 0 7.0 —	9 0 6.5 —

TABLE 3—Continued

Fasting content		5 minutes	10 minutes	15 minutes	20 minutes	30 minutes	45 minutes	60 minutes	75 minutes	90 minutes	105 minutes
Experiment 20 (Mr S) Tertiary syphilis	Total acidity	5	—	8	—	8	7	10	12	13	11
	Free HCl	0	—	0	—	0	0	0	0	0	0
	pH	7.0	—	—	—	—	—	—	—	—	—
	Chloride	63.1	—	—	—	6.5	6.5	6.0	6.0	6.0	5.5
	pP	1.7	?	—	—	—	—	34.5	—	—	35.1
Experiment 21 (Mr S)	pT	2.7	?	—	—	—	—	—	—	—	—
	Total acidity	21	—	—	—	—	—	—	—	10	12†
	Free HCl	0	—	—	—	—	—	—	—	0	0
	pH	—	—	—	—	—	—	—	—	6.0	6.0
	Chloride	52.3	—	—	—	—	—	—	—	39.5	41
	pP	2.3	?	—	—	—	—	—	—	2.3?	2.3?
	pT	2.6	?	—	—	—	—	—	—	2.2?	1.9?

* This figure was obtained from the pooled 45-, 60- and 75-minute samples

† Lavage 90 minutes

** pT = tryptic activity

made. The greatest activity was noted in the fasting content, falling somewhat thereafter. This would point to duodenal regurgitation as a regular phenomenon in cases of anacidity. How regularly this is also true in the normal is under study at present.

DISCUSSION OF PRELIMINARY OBSERVATIONS

The foregoing observations on gastric acidity in normal individuals are in accord with the findings of previous investigators. Our figures fall well within the wide limits within which free hydrochloric acid in supposedly normal individuals may vary according to Bennett and Ryle (6), Talbot (7), and Crohn and Reiss (12).

We have been particularly interested in the pH of the gastric content under various conditions. In the presence of free hydrochloric acid the pH is 3.0 or lower, and varies necessarily rather closely with the degree of free hydrochloric acid present. To the present, however, there have been very few observations on the pH of gastric contents showing no free hydrochloric acid. Under these circumstances, there may be a wide range of variation in pH—from 3.0 to 7.0 (neutrality) or above—in regard to which range ordinary determinations of gastric acidity give no information. It is noteworthy that in a small group of apparently normal individuals (about 4 per cent according to Bennett and Ryle) there is a persistent lack of free hydrochloric acid, yet these persons have no symptoms of gastric malfunction. It is conceivable that in these individuals in spite of the absence of free hydrochloric acid, the pH of their gastric contents might still be low enough (3.0 or 4.0) to be adequate (though not optimal) for peptic action. One of our anacidity patients exhibits this state of affairs. Case 17 (syphilis of the stomach) showed no free hydrochloric acid at any time. This was the only anacidity case in our series that showed reasonably good peptic activity. It was also the only case of anacidity that showed a pH lower than 5.5, the figures ranging between 3.5 and 4.0.

The only other observation on this point of which we have knowledge is that of Babbott, Johnston, Haskins and Shohl (13). It has long been known that in milk-fed infants there is usually no free hydrochloric acid in the gastric contents during the first year or so of life, yet there is good peptic activity. The above mentioned authors

studied pH variations in infants ranging in age from three to nineteen months. In the normal children the range of pH was 3.2 to 5.0, most of the figures being between 3.9 and 4.6. In our cases of true "achylia" (low peptic activity associated with achlorhydria) the pH was always 5.5 or above, most of them being between 6.0 and 7.0.

Numerous previous observations have been made on the chloride concentration in the gastric contents of normal persons and of patients with achlorhydria. Our findings in normal individuals fall within the same range as those of Wiener (14), Reisner (15) and Grund (16).

In patients with achlorhydria, which is not due to carcinoma, the chloride concentration has usually been found lower than normal. Grund and Wiener made their observations after a test meal. Strauss (17), in studying the efficiency of gastric resorption in cases of achlorhydria with pernicious anemia, found varying chloride concentrations in the fasting stomach. When the original concentration was low in the fasting content—below 40 milliequivalents—it rose progressively after the test meal to about that level or a little higher, when the concentration exceeded 80 milliequivalents in the fasting content, it fell after the test meal, through dilution. His studies were carried on over a period of forty-five minutes.

We have determined the chloride concentration in the fasting contents fourteen times in nine patients with achlorhydria. (See also tables 6, 8, 10, 15, 16 and 17.) We found fasting chloride values that uniformly fell within the normal range. After the test meal, in seven observations on six cases of achlorhydria, there was invariably a decrease in the chloride concentration, for from fifteen to sixty minutes, whereas in patients with free hydrochloric acid, there was a distinct rise in chloride concentration after the test meal. This would suggest that in the anacidity cases dilution of the stomach contents had occurred by the test meal, and, in view of the continued fall in chloride concentration, by further dilution with a fluid low in chloride content (possibly a gastric secretion). There was a late rise in chloride concentration, but in only one instance did this rise reach or exceed the previous fasting level (experiment 12).

TABLE 5
Acid given to normal individuals
 (pH and chloride figures)

Time (minutes)	Experiment 22 (case 8)				Experiment 23 (case 1)				Experiment 24 (case 2)				Experiment 25 (case 3)			
	No acid given		Acid given		No acid given		Acid given		No acid given		Acid given		No acid given		Acid given	
	pH	Cl	pH	Cl	pH	Cl	pH	Cl	pH	Cl	pH	Cl	pH	Cl	pH	Cl
1st ing	—	—	3.0	67.3	6.5	50.0	2.5	77.4	6.5	83.0	2.5	101.0	3.0	118.1	<2.0	65.0
10																
15			1.0	83.2			1.5	154.3								
20	2.0	36.6			3.0		2.0		6.0	80.0	<2.0	101.6	2.5		<2.0	109.6
30	2.0		1.5	71.1	qns				3.0		<2.0		<2.0			
40			1.5	57.5			2.0	93.7								
45	1.5	—			2.0				2.5		<2.0		2.0	79.0	<2.0	
50			qns				<2.0									
60	3.0		<2.0		2.0	53.1	<2.0		2.5	90.0	<2.0	103.7			<2.0	85.2
70			<2.0	101.2			2.0	74.8								
75	1.5	—			<2.0				2.5		<2.0		<2.0	107.9		
80																
90	2.5	102.0			2.0	81.0			<2.0	106.0	<2.0		2.0			
100																
105	1.5	—			<2.0				<2.0		<2.0	121.0			<2.0	95.4
120													2.5			
150													2.5	98.9		

EFFECTS OF HYDROCHLORIC ACID ADMINISTRATION IN NORMAL INDIVIDUALS

Four subjects of the group with normal acidity were given dilute hydrochloric acid. The amount of acid administered in each case was the maximal concentration given later to subjects with achlorhydria. It is generally accepted that the highest concentration in which hydrochloric acid occurs free in the gastric juice is about 120 to 125 millimolar, or 0.43 to 0.45 per cent (Carlson (18)). It was thought inadvisable, therefore, to exceed this concentration in the administration of acid to patients, because of possible injury to the stomach. The maximum dose of acid given to any subject was 14 cc of dilute hydrochloric acid (U S P—11.5 per cent by weight) diluted to 360 cc with water, the resultant mixture having a millimolar concentration of 120. The four normal cases accordingly received 14 cc of dilute U S P hydrochloric acid diluted to 360 cc with water. In each instance the stomach was first emptied of its fasting content, the test meal was then given and the acid mixture swallowed by the subject or introduced through the tube. Tables 4 and 5 record the results obtained, and, for ready comparison include the results obtained in the same subjects in the previous control observations.

Peptic activity was also tested in experiment 22 (case 2) and the results before and after acid administration were as follows:

Case 2 Peptic activity

	No acid given	Acid given
Fasting contents	2.8	3.3
45 minutes	3.3	
60 minutes		3.0
90 minutes	3.3	3.2

The results of maximal administration of acid in these normal individuals may be summarized as follows:

1. Total acidity was increased, the increase being greatest within the first fifteen minutes, and persisting during thirty to sixty minutes, with little difference thereafter.

2. Free hydrochloric acid was greatly increased, the increase was again most marked in the first fifteen minutes and persisted a little longer than the increase in total acidity.

TABLE 6
Case II Acid administration in anacidity (pernicious anemia)

Time (minutes)	Experiment 11 (no acid given)			Experiment 26 (7 cc. acid given in 360 cc. water)			Experiment 27 (14 cc. acid given in 360 cc. water)				Experiment 28 (14 cc. acid given in 360 cc. water)			
	Total acidity	Free HCl	pH	Total acidity	Free HCl	pH	Total acidity	Free HCl	pH	Cl	Total acidity	Free HCl	pH	Chloride
Fasting	8	0	7.0	17	0	7.0	18	0	6.0	74	11	0	7.5	53
4	—	—	—	—	—	—	—	—	—	—	44	26	2.0	38
8	—	—	—	—	—	—	—	—	—	—	58	40	<2.0	—
12	—	—	—	—	—	—	—	—	—	—	60	40	<2.0	—
15	11	0	7.0	9	0	7.0	10	0	6.5	—	—	—	—	—
16	—	—	—	—	—	—	—	—	—	—	58	44	<2.0	—
20	—	—	—	—	—	—	—	—	—	—	52	32	<2.0	—
24	—	—	—	—	—	—	—	—	—	—	28	16	<2.0	66
28	—	—	—	—	—	—	—	—	—	—	44	22	<2.0	—
30	11	0	6.5	9	0	4.0	7	0	6.5	34	—	—	<2.0	65
32	—	—	—	—	—	—	—	—	—	—	8	0	—	—
45	12	0	6.5	8	0	6.5	8	0	6.0	—	—	—	3.5	—
60	13	0	6.5	8	0	6.5	10	0	6.0	40	—	—	—	—
75	15	0	—	8	0	6.5	10	0	6.5	—	—	—	—	—
90	16	0	6.5	7	0	7.0	9	0	6.5	37	—	—	—	—
105	9	0	6.5	7	0	7.0	10	0	6.5	—	—	—	—	—

3 pH was decreased, the change tending roughly to parallel the increase in free hydrochloric acid

4 The chloride content was definitely increased during the first hour in three out of four cases studied

5 Peptic activity in the one case studied showed no change

EFFECTS OF HYDROCHLORIC ACID ADMINISTRATION IN PATIENTS WITH ACHLORHYDRIA

Dilute hydrochloric acid was administered in amounts ranging from 1 to 14 cc. Fifteen observations were made on ten patients (Eight of them had primary pernicious anemia, one syphilis of the stomach

TABLE 7
Case 11 Peptic activity

Time (minutes)	Experiment 26 (7 cc. acid given in 360 cc. water)	Experiment 27 (14 cc. acid given in 360 cc. water)	Experiment 28 (14 cc. acid given in 360 cc. water)
Fasting content	2 1	1 6	2 0
8			1 2
15	2 1		
28			2 0
30	1 3	1 6	
60	2 1	1 6	
90	2 1	2 0	
105	2 1		

and one a diaphragmatic hernia) Samples of gastric contents were removed at intervals varying from two to fifteen minutes and, in a few instances, after the withdrawal of the last sample the stomach was washed out with water and the recovered water examined

Tabulation of the results together with clinical notes on the individual cases follow

A Pernicious anemia group

Case 11 Mrs. L. H., aged 47, was admitted to the Medical Division of the University Hospital on August 25, 1924, and died February 4, 1925. Symptoms and findings on admission were characteristically those of primary pernicious anemia, with a blood count of 1,500,000 red cells. Transfusion was followed by a remission of moderate degree, the red cells reaching 3,200,000. Thereafter there was a steady decline until death. Gastric studies were made immediately after the remission, the blood count falling during the period of our observation

TABLE 8
Case 10 Acid administration in anacidity (pernicious anemia)

Time (minutes)	Experiment 10 (no acid given)			Experiment 29 (1 cc. acid given in 360 cc. water)			Experiment 30 (5 cc. acid given in 360 cc. water)			Experiment 31 (7 cc. acid given in 360 cc. water)			Experiment 32 (7 cc. acid given in 180 cc. water)		
	Total acidity	Free HCl	pH	Total acidity	Free HCl	pH	Total acidity	Free HCl	pH	Total acidity	Free HCl	pH	Total acidity	Free HCl	pH
Fasting content	25	0	6.5	—	0	—	11	0	6.0	15	0	7.5	10	0	6.5
2	—	—	—	—	—	—	—	—	—	—	—	—	60	42	2.0
4	—	—	—	—	—	—	—	—	—	—	—	—	92	82	<2.0
6	—	—	—	—	—	—	—	—	—	—	—	—	73	60	2.0
8	—	—	—	—	—	—	—	—	—	—	—	—	65	52	*
10	—	—	—	—	—	—	—	—	—	—	—	—	45	35	*
12	—	—	—	—	—	—	—	—	—	—	—	—	40	29	*
14	—	—	—	—	—	—	—	—	—	—	—	—	51	40	*
15	9	0	6.5	6	0	6.5	12	0	6.0	10	0	7.0	—	—	—
22	—	—	—	—	—	—	—	—	—	—	—	—	50	41	—
30	—	0	6.5	3	0	6.5	9	0	6.5	9	0	6.5	—	—	—
45	6	0	6.5	5	0	6.5	12	0	6.5	10	0	7.0	—	—	—
60	7	0	6.5	9	0	6.5	12	0	6.5	7	0	6.5	—	—	—
75	9	0	6.0	—	0	—	10	0	6.0	9	0	6.5	—	—	—
90	8	0	6.5	5	0	6.5	16	0	6.0	11	0	6.5	—	—	—
105	8	0	6.5	—	0	6.0	16	0	6.5	9	0	6.5	—	—	—

* Not readable

from 3,000,000 to 2,400,000. The diagnosis was confirmed at necropsy (See tables 6 and 7)

Case 10 Mr W R, aged 55 was admitted September 9 and discharged November 27, 1924. History and findings were characteristic of primary pernicious anemia, with advanced cord changes. The blood count varied from 1,700,000 to 2,900,000 red cells. Slight and transient improvement in the blood picture followed repeated transfusions. At the time of discharge he was growing rapidly worse, as shown by progressive anemia and increasing cord changes. The findings are given in tables 8 and 9.

Case 12 Mr F McC, aged 45, a patient with pernicious anemia but no cord changes, was studied from November 23 to December 3, 1924, during a period of

TABLE 9
Case 10 Peptic activity

Time (minutes)	Experiment 10 (No acid given)	Experiment 29 (1 cc. acid given in 360 cc. water)	Experiment 30 (5 cc. acid given in 360 cc. water)	Experiment 31 (7 cc. acid given in 360 cc. water)	Experiment 32 (7 cc. acid given in 180 cc. water)
Fasting content	—	—	—	1 4	1 8
15	—	1 9	—	1 4	—
22	—	—	—	—	1 8
30	—	—	1 6	1 4	—
45	—	—	1 6	1 4	—
60	1 6	2 2	1 6	1 9	—
75	—	—	1 6	1 9	—
90	—	—	1 6	—	—

remission. He had been previously admitted to the hospital on March 10, 1924, with anemia of a most advanced degree, the blood count at that time showing 500,000 red cells. Considerable benefit was derived from transfusion, but the most marked improvement followed an attack of facial erysipelas a month after admission. The blood count on May 18th was 3,200,000 red cells. He was discharged and returned November 23 for further study. During this interval he continued in fairly good health and on readmission his red cell count was 3,400,000. Treatment in the interval included the intermittent administration of dilute hydrochloric acid in 5 cc. doses with each meal. The findings in this case are shown in table 10.

Case 13 Mr J C, aged 26, was admitted to the hospital on December 2, 1924, and died February 20, 1925. He exhibited the characteristic clinical and laboratory findings of primary pernicious anemia with confirmation of the diagnosis at necropsy. The splenomegaly, increased urobilin output, and slightly

TABLE 10

Experiment 33 Case 12 Acid administration in anacidity (pernicious anemia)

Time (minutes)	No acid given				13 cc acid in 315 cc. water			
	Total acidity	Free HCl	pH	Chloride	Total acidity	Free HCl	pH	Chloride
Fasting content	7	0	6.5	42.6	11	0	7.5	37.2
4					76	70	<2.0	90.3
8					53	43	2.0	
12					47	39	<2.0	
15	7	0	6.5	34.3				
16					45	39	<2.0	
20					38	26	<2.0	68.6
24					28	18	<2.0	
28					32	18	2.0	
30	5	0	6.5					
32					26	14	3.0	
45	6	0	6.5					
47					39	16		
51					33	15		
60	12	0	7.0	68.8	22	0		69.6
75	10	0	6.5					
90	12	0	7.0	74.2				
105	11	0	6.5					

TABLE 11

Experiment 34 Case 13 Acid administration in anacidity (pernicious anemia)

Time (minutes)	No acid given				5 cc acid given in 25 cc water			
	Total acidity	Free HCl	pH	Chloride	Total acidity	Free HCl	pH	Chloride
Fasting content	15	0		56.8				
10					80	71	<2.0	74
15	15	0	6.0	40.1				
20					61	41	2.0	
30		0			51	40	<2.0	
40					41	26	<2.0	
45	8	0	6.5					
50					28	13	3.0	56.4
60	12	0	6.0					
75	12	0	6.5					
90	16	0						
105	14	0						

increased erythrocyte fragility, together with jaundice and a positive indirect Van den Bergh test pointed toward a hemolytic type of the disease. The anemia was of a severe degree and our studies were made during a period when the red cell count ranged between 730,000 and 820,000. Subsequent transfusions caused only slight transient improvement. Our findings are shown in table 11.

Case 9 Mrs S B, aged 51, was admitted to the hospital on September 19th and discharged October 20, 1924. The findings were typical of primary pernicious anemia. The blood count on admission was 2,700,000 and fell progressively to 1,800,000 just prior to her discharge. The results of our gastric studies are given in table 12.

TABLE 12

Experiment 35 Case 9 Acid administration in anacidity (pernicious anemia)

Times (minutes)	No acid given			1 cc. acid given in 560 cc. water		
	Total acidity	Free HCl	pH	Total acidity	Free HCl	pH
Fasting content	7	0	6.5	15	0	7.0
15	17	0	6.5	10	0	6.0
30	11	0	6.5	14	0	6.0
45	16	0	6.0	26	0	6.0
60	13	0	6.0	19	0	6.0
75	10	0	6.0	29	0	6.0
90	13	0	6.0	25	0	6.5
105	17	0	6.5	20	0	6.5

Case 15 Mr C H has been a very interesting patient. At the age of thirty-nine he was first admitted to the hospital on March 1, 1923, for a three weeks' stay, and has been under observation from time to time ever since. Illness had begun in 1921, and in 1923 there were characteristic phenomena of primary pernicious anemia together with paresthesias and the evidence of posterolateral sclerosis. His blood count at that time was 2,460,000 and the hemoglobin 62 per cent. Then followed a remission that has persisted to date, the blood count on April 21, 1925, was 5,020,000 and the hemoglobin 97 per cent. The patient's strength has increased, his general condition is very good, and he is at active work. While the evidence of changes in the nervous system are still present, the paresthesias have almost disappeared. Since March, 1923, this patient has been intermittently taking dilute hydrochloric acid with meals in a dosage that in the past year has been between 2 and 3 cc. The gastric studies are given in table 13. Attention is called to peptic activity which was quite low.

Case 16 Mrs. C. T., aged 44, was admitted to the hospital on March 27, 1925. The symptoms and findings were those of primary pernicious anemia, together with cord changes. The blood count on admission was 1,340,000 and the hemoglobin

TABLE 13

Experiment 36 Case 15 Acid administration in anacidity (pernicious anemia)

Time (minutes)	No acid given				8 cc. acid given in 360 cc. water				
	Total acidity	Free HCl	pH	pP*	Total acidity	Free HCl	pH	Chlorides	pT†
Fasting content	9	0 (bile)	7.5+	<1.8	12	0	7+		
15	15	0	6.5	<1.8	16	9	2.0	23.1	0
30	5	0	6.5	<1.8	15	4	2.5	26.7	0
45	8	0	6.5	<1.8	16	0	5.0		0
60	8	0	6.5	<1.8	9	0	5.5		
75	11	0	6.0	<1.8					
90	10	0	6.5	<1.8	7	0			
105									
120					4	0	7.0	12	

* pP = peptic activity

† pT = tryptic activity

TABLE 14

Experiment 37 Case 16 Acid administration in anacidity (pernicious anemia)

Time (minutes)	No acid given		8 cc. acid given in 360 cc. water			
	Total acidity	Free HCl	Total acidity	Free HCl	pH	pT*
Fasting content	8	0	8	0	7.5	+
15	18	0	12	0	4.0	
30	16	0	12	0	4.0	
45	q n s	0	12	0	5.0	0
60	q n s	0	12	0	5.0	
75	8	0				
90	8	0	10	0	6.5	
105	12	0	8	0	6.5	

* pT = tryptic activity

30 per cent. The patient's condition has been little influenced by transfusion and at the time of this writing she is becoming steadily worse. No control studies were made on pH, chlorides or ferments. The results of the gastric studies are stated in table 14.

Case 21 Mr J McC, aged 48, was admitted to the hospital on April 9, 1925. He dates his illness from July 1922, when he gave blood for a transfusion to his sister, who later died of primary pernicious anemia. The patient's symptoms and findings on admission were those of primary pernicious anemia of an advanced degree. The blood count has fluctuated between 670,000 and 1,250,000 and the hemoglobin between 21 and 30 per cent, a slight improvement following two transfusions. The patient's condition was so serious that only a single gastric study was made after acid administration and there are therefore no control figures. The findings are given in table 15.

Five other patients with pernicious anemia were given 1 cc each of dilute hydrochloric acid. In no instance was free hydrochloric acid found in the stomach on fractional analysis. As pH, chloride and ferment studies were not made in these cases, we have omitted them from this report.

TABLE 15

Experiment 38 Case 21 Acid administration in anacidity (pernicious anemia) Four cubic centimeters acid given in 180 cc water (No control observations)

Time (minutes)	Total acidity	Free HCl	pH	Chloride	pT*
Fasting content	5	0	7.5+	30.2	2.9
15	14	5	3.0	22.6	1.9
30	13	5	3.0	24.5	1.9
45	13	0	4.0		
60	6	0	6.5	33.5	1.9
75	7	0	6.5		
90	8	0	6.5		
105	7	0	6.5		

* pT = tryptic activity

B Non-anemic group

Case 17 Miss E G, aged 22, was admitted to the hospital on October 22d and discharged December 31, 1924. There was a history of indigestion and pain after meals at the onset six months before, followed by signs of increasing pyloric obstruction with vomiting, constipation and loss of weight. There were stigmata of syphilis, possibly congenital, in the form of old vitreous opacities and corneal changes, widespread choroiditis, and a general lymphadenopathy. The blood Wassermann on repeated occasions was strongly positive. The blood count was practically normal. Gastro-intestinal x-ray showed an organic obstruction at the pylorus, which was considered by Dr. Pancoast to be probably syphilis of the stomach, but with the possibility of carcinoma. She was given active anti-syphilitic treatment, including salvarsan and iodide, with marked clinical im-

provement When last seen (April 1, 1925) she was symptom-free, her weight had increased from 81 pounds to 131 pounds, and fluoroscopic examination of the stomach showed definite improvement of the local lesion, which was still present

TABLE 16
Experiment 39 Case 17 Acid administration in anacidity (gastric syphilis)

Time (minutes)	No acid given					14 cc acid given in 360 cc. water				
	Total acidity	Free HCl	pH	Chlorid	pP*	Total acidity	Free HCl	pH	Chloride	pP*
Fasting content	60	0	3 0	63	3 2	6	0	7 5	44	2 4
15	64	0	3 0	62 1		11	2	3 0	47	2 4
30	30	0	3 0			20	8	3 0	47	2 4
45	28	0	3 5			16	2	4 0		
60	26	0	3 5	38 9	3 2	18	0	4 0		2 4
75	26	0	3 5			17	0	4 0		
90	30	0	3 5			16	0	4 0		1 9
105	50	0	3 5	52 7		17	0	4 0	65	

* pP = peptic activity

TABLE 17
Experiment 40 Case 19 Acid administration in anacidity (idiopathic)

Time (minutes)	No acid given				7 cc acid in 360 cc water			
	Total acidity	Free HCl	pH	Chlorid	Total acidity	Free HCl	pH	Chloride
Fasting content	16	0	7 0	74 6	9	0	7 0	77 4
10					67	54	1 5	83 2
15	11	0	6 5	70 7				
20					60	50	2 0	
30	9	0	6 5		46	39	2 0	
40					32	17	2 5	60 2
45	8	0	7 5	52 0				
60	12	0	7 0	58 3				
75	9	0	7 0					
90	7	0	7 0					
105	9	0	6 5					

but no longer obstructive The diagnosis of syphilis of the stomach seems amply warranted Our observations were made in the second week of her stay in the hospital The findings are given in table 16

Case 19. Mr J E, aged 54, was in the hospital from November 17th to December 23, 1924. There was a long history of constipation, for which an operation had been performed nine years previously, and later a second operation for supposed postoperative adhesions. Gastro-intestinal X-ray revealed a hernia of the stomach through the diaphragm, a finding which was confirmed by esophagoscopy. There was a persistent achlorhydria. Our findings are given in table 17.

ANALYSIS OF TABLES VI TO XVII INCLUSIVE

1 *a* Free hydrochloric acid did not appear in the gastric contents after the administration of 1 cc of dilute acid

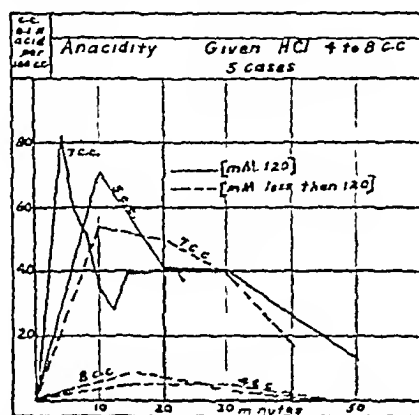


FIG 1 CURVES OF FREE HCl IN CASES NUMBERS 10, 13, 15, 19 AND 21 AFTER RECEIVING 4 TO 8 CC OF ACID

b The administration of 4 to 8 cc of acid was followed in two cases (primary pernicious anemia, diaphragmatic hernia of the stomach) by an immediate rise of free hydrochloric acid to 71 and 54 milliequivalents respectively after ten minutes, free acid continuing to be present in diminishing amounts up to fifty minutes. A third case (primary pernicious anemia) showed free acid at two minute intervals for twenty-two minutes, the maximum being 82 milliequivalents at four minutes. Two other cases of primary pernicious anemia showed free acid at fifteen and thirty minutes. The curves are shown in figure 1. Three cases (primary pernicious anemia) showed no free acid on fifteen-minute testing. The concentration of the acid given

to these patients was less than 120 millimolar in seven instances, and four of these failed to show free acid. In two instances the concentration of the acid given equaled 120 millimolar, and both of these patients showed free acid.

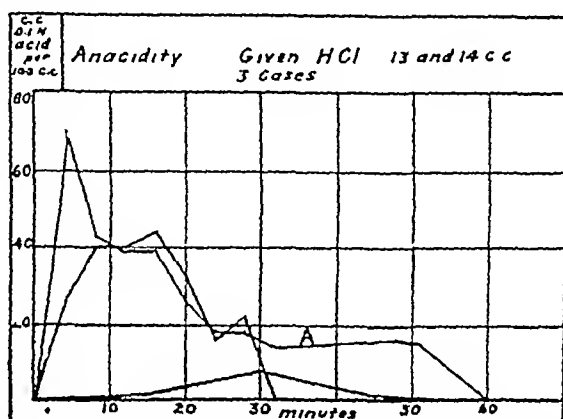


FIG 2 CURVES OF FREE HCl IN CASES NUMBERS 11, 12 AND 19 AFTER RECEIVING 13 TO 14 CC OF ACID

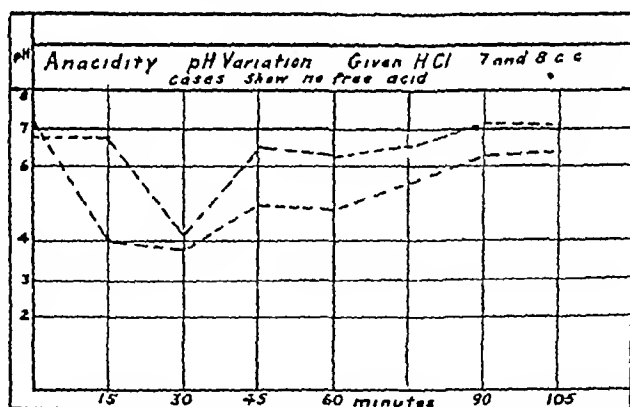


FIG 3 pH CURVES IN CASES NUMBERS 11 AND 16 AFTER RECEIVING 7 AND 8 CC OF ACID RESPECTIVELY

c The administration of 13 to 14 cc of acid (millimolar concentration 120) in four instances was followed three times by the appearance of free acid, lasting from twenty-eight to fifty-one minutes (see fig 2). The peak of the curve in two cases (primary pernicious anemia) occurred in the first specimen that was removed. A case of gastric syphilis showed a very slight rise in the free-acid curve. One

case of pernicious anemia which showed free hydrochloric acid for twenty-eight minutes when the acid administered was given through the stomach tube and specimens removed every four minutes (curve A, fig 2), failed to show free acid when the same amount of dilute acid was swallowed and specimens were removed every fifteen minutes

2 Total acidity changes following acid administration roughly paralleled the changes in free hydrochloric acid

3 The effects on the pH may be summarized as follows When free hydrochloric acid was present after the administration of acid, the pH fell to 3.0 or below, the degree of change being roughly proportional to the degree of free hydrochloric acid present When there was no free hydrochloric acid present, the pH in most instances was unchanged In two cases, however, there was a fall in pH to values about 4.0 as shown in figure 3 This type of change was never observed after the administration of less than 4 cc of acid

4 The results of our studies on the comparative chloride values do not lead to any definite conclusions The most constant change was an immediate increase after acid administration, comparable to the increases in acidity

5 In no instance was any effect on peptic activity noted

6 Trypsin was present in the fasting contents of all cases studied except one, and tryptic activity was but little less than that in duodenal contents In the one exception the material obtained from the fasting stomach was chiefly mucus

DISCUSSION OF EFFECTS OF HYDROCHLORIC ACID ADMINISTRATION IN ACHLORHYDRIA

The effects of administration of hydrochloric acid on the various types of achlorhydria have been repeatedly studied in the past In the pernicious anemia group the observations have been limited to the changes occurring in free and total acidity, and in bacteriacidal activity, with the general conclusion (Leo, Crohn, Shaw—previously cited) that 1 cc of dilute hydrochloric acid produced no effect, and that it usually required at least 3 cc to cause the appearance of free hydrochloric acid after a test meal In carcinoma of the stomach, Fischer (19) found that larger amounts of hydrochloric acid were necessary to produce free acid in the gastric contents than were

required in other types of achlorhydria. This he attributed to the buffer effect of protein split-products. In the cancer group the chlorides have also been studied, with the finding usually of higher values than in other forms of achlorhydria.

Little or no attention has been paid by previous observers to possible changes in the pH of the gastric contents in achlorhydria after the administration of hydrochloric acid. While the presence of free hydrochloric acid indicates a pH of 3.0 or below, yet a wide range of pH variation is possible in achlorhydria (from 3.0 to 7.0 or above), a range in which changes are of great importance in connection with peptic activity. It seemed to us possible that the administration of hydrochloric acid, although insufficient to produce free hydrochloric acid in the stomach contents, might nevertheless lower the pH to a range between 4.0 and 3.0. Such a pH, even though not optimal, would materially enhance peptic activity.

In our experience, small doses of acid (1 cc) not only failed to produce free hydrochloric acid, but also did not appreciably lower the pH. In an occasional case larger doses did cause a fall in pH, but not to the point of appearance of free hydrochloric acid. It is conceivable, therefore, that in cases of anacidity gastric physiology may be favorably influenced at times by hydrochloric acid administration without the appearance of free acid, in that the action of the pyloric sphincter may be affected. The administration of hydrochloric acid in our series, however, did not in any instance increase peptic activity measured at optimal pH, in other words, it did not stimulate pepsin secretion or prevent its destruction. Nor is this true only after single doses of acid. Cases 12 and 15 have been receiving from 2 to 4 cc of dilute hydrochloric acid fairly constantly for nine and twelve months respectively, yet in neither instance was peptic activity at optimal pH greater at the end of that time than when treatment was begun. It would seem, therefore, that in primary pernicious anemia the diminished power to secrete pepsin cannot be restored by this therapy. A therapeutic corollary would be the administration routinely of pepsin as well as hydrochloric acid in attempting to reproduce normal gastric conditions.

The appearance of free hydrochloric acid after acid administration seems to depend on two factors (a) the nature and severity of the

underlying cause of the achlorhydria, and (b) the amount and concentration of acid given. In general, our best results were obtained in cases of anacidity that were not due to primary pernicious anemia, and in cases of primary pernicious anemia in a state of remission. In the more amenable cases, 4 to 8 cc of dilute acid, usually in a 120 millimolar concentration, produced free hydrochloric acid, in the severest cases, 14 cc of acid at times failed or produced only transient free hydrochloric acid.

When free acid does appear after the administration of hydrochloric acid, the maximum concentration is found in the first specimen removed, and falls progressively thereafter. This is true both after single doses and after prolonged administration of acid. There is no evidence, therefore, to show that acid administration in achlorhydria is followed by increased secretion of acid by the gastric mucosa.

It was found quite feasible to administer doses of 4 to 8 cc of acid over long periods of time. Given in tea or buttermilk and sipped during and after the meal, this amount of acid is easily taken by the patient. Larger doses are impracticable.

THE FATE OF ORALLY ADMINISTERED HYDROCHLORIC ACID IN ACHLORHYDRIA

It is remarkable with what rapidity hydrochloric acid, when administered orally, disappears or is neutralized in the gastric contents in patients with achlorhydria. A number of factors must be considered: neutralization by an alkaline gastric secretion, regurgitation of alkaline duodenal contents, swallowed saliva and mucus, absorption of acid from the stomach, and rapid passage onward into the duodenum. Our studies of the chloride concentration have shed no light on the problem. The constant presence of trypsin in the fasting gastric content proves that duodenal regurgitation at least plays a part.

CONCLUSIONS

- 1 The pH of the gastric content in achlorhydria may range from 3.0 to 7.0+.
- 2 In our cases of primary pernicious anemia the pH was always 5.5 or higher—levels at which peptic activity is markedly depressed.

3 In achlorhydria, not associated with primary pernicious anemia, the pH may in some cases be between 3.0 and 4.0, levels which permit of considerable (if not optimal) peptic activity, and consequently a fairly efficient gastric function

4 The presence of trypsin in the fasting content of the stomach in achlorhydria points to duodenal regurgitation as a constant phenomenon in this condition

5 The administration of dilute hydrochloric acid in cases of achlorhydria in a dosage of 1 cc is not followed by the appearance of free hydrochloric acid and fails to produce any material change in the pH of the stomach contents

6 After larger doses (4 to 14 cc) the pH of the gastric contents is lowered in some instances below 3.0 with the presence of the usual test for free hydrochloric acid, in others not below 3.0, and therefore not showing the usual test for free acid. In the latter case the change may nevertheless be sufficient to influence gastric function

7 The effect produced by the acid varies with the nature and severity of the disease underlying the achlorhydria, and with the concentration of the acid administered

8 In the treatment of achlorhydria doses of 4 to 8 cc of dilute hydrochloric acid have been found practicable, adequate, and preferable to the usual dose of 1 cc

9 The administration of hydrochloric acid, even though long continued, does not stimulate the secretion of pepsin or effect the spontaneous appearance of free hydrochloric acid in the stomachs of patients with achlorhydria

10 In two cases of primary pernicious anemia remissions of nine and twelve months have co-existed with the continued administration of dilute hydrochloric acid in doses of 4 cc. Such acid therapy is worthy of further observation

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BLOOD SUGAR TIME CURVES

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(Received for publication May 21 1926)

Blood sugar time curves, or what are more generally, but less exactly, termed sugar tolerance curves, are now receiving wide clinical application. As a result of this, various disorders, other than diabetes mellitus, are known to result in, or be associated with, a disturbance in the metabolism of carbohydrates. The response of the individual to glucose ingestion is probably the best of indices of such a disturbance.

It is obvious that the "normal" type of curve must be recognized in order to detect pathological variations. Within the last few years, many different results, obtained in apparently normal individuals, have been recorded. A study of the literature however, shows that in obtaining these results, not only have various procedures been followed as to selection of the amount and type of sugar, and the periodic collection of blood and urine specimens but the analytical methods for blood sugar estimation have been different. The literature on the subject has become voluminous, and for brevity no attempt at review will be made here, since it would serve no particular purpose in the present discussion.

In spite of the many procedures and different technical methods, a statistical study of all the results would tend to give a fairly approximate idea of what is the response of the normal individual to glucose ingestion. This view is based upon one of the theorems of statistics, namely, that the arithmetical mean of a large series of observed values, is the most probable value of the quantity measured. Such painstaking study has, recently, been made (1). In a comprehensive study, including a statistical analysis of 4000 individual blood sugar determinations in 971 cases, Gray arrived at standards which are generally accepted. The values of the mean and the mode (the most frequent

results) of the series agreed very closely. The blood sugar time curve is characteristic. The fasting blood sugar is about 0.090 per cent. Following the administration of glucose, the maximum increment is noted within 30 minutes, and the value is below 0.180 per cent, the average peak being 0.140 per cent. The rate of decline is at first rapid, then slower. Within three hours the blood sugar concentration is equal to or below that of the fasting period.

In only 10 of 431 cases was the fasting level above 0.120 per cent. Eight of these were 0.130 per cent. In one case it was 0.150 and in another 0.160 per cent. These data are of particular interest since, in the majority of cases (300 out of 479), the dose of sugar administered by the various workers was that now generally employed, namely, 70 to 100 grams. In the majority of the 300 cases abnormal glycosure-sis did not occur. It was, however, found in a sufficient number, namely 129, to emphasize the difficulty of the interpretation of this phenomenon.

The majority of observers are also in agreement as to the relation between the type of the curve and the dose of sugar given. With as little as 5 grams an appreciable increase was noted. The highest peak with the smallest dose was as great as the average peak with the larger dose. With doses exceeding 25 grams the peak was not affected. Increasing the dose merely prolonged the time necessary for the blood sugar to return to its original fasting level. The difference between the effects of the starches and glucose was too small to determine any special preference.

In spite of these findings, however, one frequently meets with differences of opinion as to what may be regarded as a "normal" curve. There appears to be a fairly general agreement as to the value of the concentration of sugar in the blood in the post absorptive state and the time necessary for the blood sugar to return to the basal level. There is, however, much difference of opinion as to the maximum concentration of blood sugar which may be regarded as normal following glucose ingestion. Blood sugars as high as 0.250 per cent have not been regarded by some observers as pathological, providing they have returned to the normal level in two and a half or three hours. These high values have been attributed to the great differences, in different individuals, of rates of absorption of glucose from the alimentary canal.

It appears reasonable that this problem would be much simplified if a large series of observations were made by one procedure and one technical method of blood sugar estimation. This has been done and the object of this communication is to record the results of over seven hundred such blood sugar time curves. In each case the following routine was adopted. It is one generally in use, but it is described in detail for the purpose of the present discussion.

No food nor fluids of any kind were allowed after 6 p.m. the evening before the test. In the morning, the patient voided. One hundred grams of ordinary glucose,¹ dissolved in 250 cc. water and flavored with lemon juice were then given, and blood and urine specimens were collected 30, 60, 120 and 180 minutes later.² Benedict's qualitative test was used for detecting sugar in the urine. All blood sugar estimations were made by the Lewis-Benedict method, with slight modifications, but with no alteration in principle. A brief remark with reference to this method may not be out of place.

It is a generally recognized fact, that this procedure yields higher values than the Folin-Wu method. In an investigation such as this, there are, however, other and equally important considerations in the selection of a method. It might be pointed out that much of our knowledge of blood sugar time curves and our ideas of fasting and threshold blood sugar values are the results of work done with picric acid procedures. In fact, though the prevalent method is probably that of Folin and Wu, there is relatively little in the literature on studies of fasting and threshold values made with the use of the Folin and Wu method, and with "ordinary" glucose. It appears rather illogical to obtain blood sugar time curves by one method, and in the interpretation of results, accept as standards, values obtained by another method, when the methods do not yield identical values.

¹ The term ordinary glucose refers to the commercial product. The chemically pure sugar was not used, since it is rarely employed for clinical purposes, and the value of data obtained with it would therefore be limited for comparative purposes.

² The collections of accurately timed blood specimens were made possible with the assistance of Drs. F. G. Stearn, Harold Eberts, C. N. Ramsay, P. Ward, E. S. Mills, I. Trossman, M. C. Childs, J. B. Ross, F. Hume and B. Alexander, who at different times were house physicians in the Metabolism Service. Grateful acknowledgment is due the Nursing Staff in charge of the various wards who were responsible for the collection of urinary specimens.

The actual glucose content of human blood is still unknown. No better demonstration of this fact can be found than in the continual researches and critical analyses of each other's work by Folin and Benedict. The more recent methods tend to yield lower values. While this important problem is still under investigation it is considered advisable to adhere to one method, the technique of which has been thoroughly mastered. This has been done in this laboratory and during the last six years approximately 30,000 determinations have been made with the method employed in this study. As a result of this experience, we are quite in agreement with Benedict (2) in his assertion that rarely has the clinician been misled in the interpretation of blood sugar results obtained by the picric acid method. We have also frequently noted the fact, mentioned by Benedict, that the rate of return of the blood sugar to the normal level, as found with the picric acid method, paralleled the clinical progress of diabetics more closely than the results with the Folin and Wu method, with which the decline in blood sugar concentration was more rapid.

The method of investigation was, firstly, to accept as a standard curve, for statistical purposes, the average values found by Gray for normal individuals, which are as follows

- a* Post absorptive blood sugar, 0.080 to 0.120 per cent
- b* Maximum blood sugar following glucose ingestion not above 0.180 per cent
- c* Blood sugar at the end of 3 hours 0.080 to 0.120 per cent

The clinical picture in each case was correlated with the blood sugar time curve.

Evidence suggesting diminished sugar tolerance was found in 268 out of 713 cases. In 140 out of the 268 cases, the curves showed that hyperglycemia was present at the end of three hours. These are excluded from the present discussion. We are particularly concerned with the 128 remaining curves which showed maximum blood sugar concentrations above 0.180 per cent, and normal blood sugars at the end of 180 minutes. These were divided into two groups according to the clinical pictures with which they were associated as follows

Group 1 Those conditions known to cause, or to be associated with a disturbance of carbohydrate metabolism

Group 2 Those in which no reason could be found for the abnormalities in the curves

All subjects were hospital cases. In table 1 are recorded in each group the different clinical conditions, and their incidence. It will be noted that in 99 (group 1) out of the 128 cases an incidence of 77.3 per cent—the clinical conditions were such as to offer reasonable explanations for the characters of the blood sugar time curves. In only 29 cases

TABLE 1

Subjects with maximum blood sugar concentrations above 0.180 per cent and normal blood sugar after 180 minutes

Group 1		Group 2	
Clinical diagnosis	Incidence	Clinical diagnosis	Incidence
	<i>cases</i>		<i>cases</i>
Glycosuria*	24	Gastric ulcer	2
Exophthalmic goiter	21	Migraine	2
Toxic adenoma	4	Chronic tonsillitis	1
Rheumatoid arthritis	8	Menopause	4
Chronic pancreatitis	3	Tabes	1
Chronic cholecystitis	5	Chronic pleurisy	1
Cholelithiasis	3	Myocarditis	5
Hepatic cirrhosis	8	Tuberculosis	5
Convalescent from catarrhal jaundice	1	Angina pectoris	1
Chronic eczema†	11	Chronic endocarditis	2
Pregnancy	1	No disease, chronic	
Chronic nephritis	8	Outdoor visitor	1
Hemiplegia	1	Chronic bronchitis	1
Encephalitis lethargica	1	Duodenal ulcer	2
		Chorea	1
Total	99	Total	29

* All of these subjects came under observation because glycosuria was discovered during examination for life assurance policies. In fifteen of these there was a family history of diabetes.

† The Department of Dermatology is now making an investigation of this type of case. Some of these subjects have been noted to improve on low carbohydrate diets. In two of these there was a family history of diabetes.

(group 2) did there appear to be insufficient reason for the high blood sugar noted.

A detailed analysis of the maximum blood sugar concentrations following glucose ingestion are recorded in table 2. It is obvious from a glance at this table that the highest blood sugar values were found in group 1. Of the 29 cases in group 2, 19 had blood sugars below 0.200 per cent. These results may be stated differently. Of 713 subjects

only 29 showed hyperglycemias (blood sugars above 0 180 per cent) following ingestion of ordinary glucose that could not be attributed to disease. In 19 of these the blood sugars ranged between 0 181 and 0 200 per cent. Marked hyperglycemias (blood sugars above 0 200 per cent) were therefore found in only 10 out of 713 cases, an incidence

TABLE 2

Cases from table 1 analyzed with respect to maximum blood sugar concentration

Maximum blood sugar <i>per cent</i>	Incidence	
	Group 1	Group 2
	<i>cases</i>	<i>cases</i>
0 181-0 185	11	8
0 186-0 190	12	4
0 191-0 195	4	5
0 196-0 200	17	2
0 201-0 205	6	3
0 206-0 210	2	1
0 221-0 215	3	0
0 216-0 220	0	1
0 221-0 225	16	0
0 226-0 230	4	1
0 231-0 235	10	1
0 236-0 240	0	0
0 241-0 245	3	0
0 246-0 250	7	1
0 251-0 255	0	0
0 256-0 260	0	1
0 261-0 265	0	0
0 266-0 270	2	1
0 271-0 275	0	0
0 276-0 280	0	0
0 281-0 285	0	0
0 286-0 290	0	0
0 291-0 295	1	0
0 296-0 300	0	0
0 301-	1	0

of 1 4 per cent. These data therefore appear to offer reasonable proof that in the great majority of instances, blood sugar values above 0 180 per cent are pathological. As to blood sugars above 0 200 per cent, the percentage incidence of these, in individuals with no apparent conditions to account for disturbance in carbohydrate metabolism, was so small that it would be practically necessary to assume an infallible technique to regard their occurrence as positive.

Further evidence that maximum blood sugar concentrations above 0 180 per cent following ingestion of "ordinary" glucose are distinctly pathological was found by correlating post absorptive blood sugar values and blood sugar time curves. Of the 713 subjects, 103 had hyperglycemia (above 0 120 per cent) in the post absorptive state. These subjects were grouped according to their post absorptive blood sugars, 0 120 to 0 125 per cent, 0 126 to 0 130 per cent, etc. In table 3 are recorded in order the different groups, the number of individuals in each group, and the number of these who had diminished sugar tolerance. These results rather strikingly demonstrate the real significance of blood sugar time curves, in which maximum

TABLE 3

Correlation of high post-absorption blood sugar value with occurrence of diminished tolerance in blood sugar time curve

Post absorptive blood sugars	Number of cases	Diminished tolerance	
		Number of cases	Percentage incidence
<i>per cent</i>			
0 121-0 125	23	14	60 8
0 126-0 130	23	18	78 2
0 131-0 135	9	8	88 8
0 136-0 140	14	13	92 8
0 141-0 145	7	7	100 0
0 146-0 150	6	6	100 0
0 151-	21	21	100 0

sugar concentrations reach above 0 180 per cent. In no case with a basal blood sugar of 0 140 per cent or over, did the curves fail to rise above the 0 180 per cent level. In spite of the fact that the slight differences of values between 0 121 to 0 125 per cent may easily have been the results of technical errors, over sixty per cent of this group showed diminished tolerance in their curves. These results further corroborate the view expressed by Benedict that blood sugar values determined by the picric acid method have rarely misled the clinician in his interpretation.

An attempt was made to correlate glycosuria with the character of the blood sugar curve. Glycosuria was found in 309 out of the 713 cases—an incidence of 43 3 per cent. This agrees very closely with that found by Gray in his study. As stated above Gray found glycosuria in 129 out of 300 cases—an incidence of 43 per cent. Of our 268

cases showing positive curves, 200 showed glycosuria. It will therefore be seen that 109 subjects with normal blood sugar curves had glycosuria. The deduction made from these observations is that glycosuria following ingestion of "ordinary" glucose is not a reliable clinical index of a disturbance of carbohydrate metabolism.

SUMMARY

A large number of blood sugar time curves were obtained in order to determine whether individuals, who have maximum blood sugar above 0.180 per cent, following ingestion of ordinary glucose, but whose blood sugar returns to the normal level at the end of three hours, may be regarded as normal.

By correlating the blood sugar time curves with the clinical pictures it was found, in the great majority of cases, that maximum concentrations above 0.180 per cent were associated with conditions known to cause or be associated with a disturbance of carbohydrate metabolism.

In view of these findings, it is regarded that the presence of a normal blood sugar three hours after glucose ingestion is not a sufficient basis for the diagnosis of a normal carbohydrate metabolism.

A blood sugar increase of over 0.180 under the same set of conditions, should, in the majority of cases, be regarded as pathological. This figure becomes more significant in view of the fact that the prevalent method of determining blood sugar (Folin and Wu), yields, as a rule, values lower than the picric acid method.³

This work was done with the technical assistance of Miss Althea B. Frith, Miss Jane Spier and Miss Helen McGachen.

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³ While this paper was in the press an article by Petty & Stoner appeared in the American Journal of Medical Sciences (1926, *clxxi*, 42), on "Respiratory quotient curves in the diagnosis of diabetes." By respiratory metabolism data these workers came to the same conclusion as we have, namely, that a number of individuals whose blood sugar was normal before and three hours after the administration of glucose were shown to be definitely diabetic, and that such blood sugar findings do not form a sufficient basis for the exclusion of the diagnosis of diabetes.

GUANIDINE EXCRETION IN RELATION TO HYPERTENSION

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(Received for publication May 11, 1925)

A series of observations have been made recently, the results of which suggest, a relationship between arterial hypertension and guanidine metabolism. Major and Stephenson (1) studying the influence of certain urinary constituents on blood pressure, noted that methyl guanidine, a product of normal metabolism and a constituent of normal urine, had marked pressor effects. These authors suggested that this compound, if produced in excess or imperfectly excreted, might account for increased blood pressure. Creatinine and creatine, substances closely allied to methyl guanidine chemically, were found inert. Later (2) observations confirmed the rise in blood pressure following experimental injection of methyl guanidine. In addition it was then noted that, in short period experiments, rise in blood pressure produced by guanidine compounds could be abolished by the use of calcium or potassium chloride. The latter is of interest, in view of the clinical results obtained from the use of these salts as reported by Addison (3) and Addison and Clark (4), who found that these inorganic salts, when given in large doses (about 15 grams per day) over long periods of time, produced a diuresis and accompanying this there was a decided and prolonged fall in blood pressure in a large percentage of cases of hypertension. In a further communication Major (5) reported two cases of arterial hypertension in which the daily excretions of guanidine bases were observed. In both, during produced diuresis, a rise in urinary excretion of dimethyl-guanidine was accompanied by a fall in blood pressure. Experimental evidence was also presented by the same worker (6) favoring the view that the elevation in blood pressure

following administration of guanidine compounds is due to the peripheral constriction of the arterioles

In view of the above findings we have made a study of the excretion of guanidine compounds in a series of normal individuals and in patients with hypertension

In all their investigations Major and his co-workers employed the technical method described by Findlay and Sharpe (7) for the quantitative estimation of dimethylguanidine. In our earlier work with this technique some difficulty was encountered in that, in spite of the theoretical differences in solubility between guanidine picrate and the creatinine compound, the latter was found to interfere greatly with quantitative results. The method employed, subsequently, was that more recently described by Sharpe (8). The steps suggested by Sharpe for the separation of creatinine from guanidine (fractional crystallization or the use of blood charcoal) have yielded results sufficiently quantitative for the present purpose. By this method Sharpe has shown that about 90 per cent of added guanidine hydrochloride can be recovered from a solution containing this compound, sodium chloride, potassium chloride and creatinine hydrochloride. The same percentage recovery may also be noted when guanidine hydrochloride is added to normal urine. The writers are indebted to Dr Sharpe for the helpful suggestions made in a personal communication, with reference to this method.

Observations were made first upon a series of normal urines. As noted by Major, marked fluctuations were found in the amounts excreted daily by the same individuals. Two possible explanations for this are suggested. Like most of the other nitrogenous substances excreted, the daily amounts may vary. In view, however, of the constancy of creatinine excretion and the chemical relationship between creatinine and methyl guanidine, it is quite possible that the total amount of guanidine bases excreted daily is constant for the individual, but that the relative proportions of the different compounds vary and that all of these are not recovered quantitatively by the method employed. In view of the above consideration data were collected to determine the average daily excretions in normal individuals. In each case observations were made on three successive days. The results on ten such individuals are recorded in table 1.

In table 2 are shown the average daily excretions in thirteen cases of marked arterial hypertension. In each case are also recorded, in order, the values of the blood urea nitrogen, blood creatinine, urine urea concentration following ingestion of 15 grams urea, and the urea concentration factor $\left(\frac{\text{mgm Urea N per 100 cc urine}}{\text{mgm Urea N per 100 cc blood}} \right)$, the systolic and diastolic blood pressures, and the presence or absence of albumin and casts in the urine. Practically all blood pressure observations were made by the same individual, and the beginning of the fourth phase was accepted as the diastolic pressure.

DISCUSSION

The average daily excretion of dimethyl-guanidine in the thirteen cases of hypertension was 105 mgm. This was lower than that found in the normal subjects, namely 119 mgm, but equal to that found by Major in normal subjects. Since with our present knowledge the variables affecting excretion of guanidine bases are not known one cannot accurately determine which deviations from the "means" are purely fortuitous. Application of statistical methods to the phenomenon of variation observed here would therefore yield information of limited significance. It is merely stated that a glance at both tables 1 and 2, shows that in the majority of instances the low average values of guanidine excretion are associated with the high values of the diastolic blood pressures. This is of interest in view of the suggested mechanism by means of which the guanidine compounds raise the blood pressure, namely, by peripheral constriction of the arterioles.

Daily observations were made in one case over a period of two weeks. The subject, a female, aged 35 years, was admitted into the Medical Service of one of the writers (C. P. H.) on October 6, 1925. The clinical picture was that of chronic interstitial nephritis, associated with syphilis. There was marked thickening of the blood vessels, cardiac hypertrophy, and some retinal changes. Blood pressure systolic 250, diastolic 160. The urine contained albumin and casts. The blood urea nitrogen was 14 mgm per 100 cc blood, creatinine 1.20 mgm per 100 cc. The urea concentration factor was 16.

The patient was kept in bed throughout the period of observation and on a constant diet. Daily examinations were made with reference

to blood pressure and the excretion of water and guanidine. When the blood pressure tended to remain at a constant level following the initial fall due to rest in bed, calcium chloride was given in doses of 5 grams three times a day. The above daily examinations were continued. The results are recorded in table 3. It will be noted that during the first four days, though the systolic blood pressure fell from 250 to 220 mm and the diastolic pressure fell from 160 to 125 mm there was no increase in the excretion of guanidine. This was con-

TABLE 3

Blood pressure, excretion of urine and dimethyl-guanidine in a case of chronic interstitial nephritis (Case 4742-25)

Date	Blood pressure		Daily excretion of dimethyl- guanidine	Volume of urine
	Systolic	Diastolic		
			mgm	cc
October 6	250	160	74	950
October 7	205	130	45	1,400
October 8	220	130	82	870
October 9	220	125	68	920
October 10*	225	130	112	2,400
October 11	210	135	386	3,550
October 12	185	125	240	2,620
October 13	180	120	318	3,000
October 14			194	2,990
October 15			206	3,900
October 16	178	122	370	2,860
October 17	180	118	385	2,800
October 18	170	115	294	3,100
October 19	180	110	192	1,660

* Calcium chloride treatment (5 grams t.i.d.) commenced.

trary to that expected if there is perfect correlation between guanidine excretion and blood pressure. During this period *there was no polyuria*. Following the institution of the calcium chloride treatment (October 10) both the systolic and diastolic pressure fell still further. *This was accompanied by a marked diuresis*, and the excretion of guanidine bases was increased. Urine volume output therefore appeared to influence excretion in this case. The fact is merely mentioned that following calcium chloride treatment a lowering of the blood pressure occurred. It does not necessarily follow that this was caused by the

treatment As a matter of fact we have sufficient data proving the inefficacy of this therapeutic measure

In view of all the above findings, the relationship between arterial hypertension and decreased guanidine excretion is suggestive, and on the whole, there is some corroboration of the work of Major and his associates, on the relation between increased guanidine excretion and diuresis However, the fact that some individuals with marked hypertension excrete normal amounts of guanidine bases, and that in the case reported above (No 4742) the fall in blood pressure during the earlier period of observation was not accompanied by an alteration in the excretion of these substances, warns us that more must be known of this relationship

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PROCEEDINGS OF THE EIGHTEENTH ANNUAL MEETING
OF THE AMERICAN SOCIETY FOR CLINICAL INVESTIGATION
HELD IN ATLANTIC CITY, NEW JERSEY, MAY
3, 1926

Dr Albion Walter Hewlett

Born 1874 Died 1925

During the past year we have lost by death one of the small group of men to whom the foundation of this society was due and one who later became its President, Dr Albion Walter Hewlett

While still a medical student his intellectual ability, his industry and his power of clear thinking and accurate expression marked him as one who was destined to become an important figure in his profession

He very early began to exhibit an interest in the fundamental processes concerned in disease, and was not content merely to observe and describe the superficial and obvious This interest led him to the study of normal physiological processes and to apply the methods of this study to the sick

These interests, which twenty years ago were not so common as they are now, led him and others of like mind to gravitate together, and as a result there came into being this organization of men bound together for searching out the underlying secrets of disease

But Dr Hewlett was also interested in men, in sick individuals, in the classification of disease, in diagnosis and in treatment He was a skillful clinician These qualities were the ideal ones for the successful teacher and leader, and it was not surprising therefore that he was called, while very young, to become the Professor of Medicine at one of the important universities, the University of Michigan

While at Ann Arbor, Dr Hewlett endeared himself to faculty and student body alike His sincerity and the thoroughness with which he executed the trust placed in him as head of the department of medicine was an inspiration and stimulation to all, particularly to the young

men who were fortunate enough to become intimately associated with him

Under Dr Hewlett's direction the Department of Medicine at Michigan continued to grow and made marked and well recognized advancement Under his leadership valuable contributions to scientific medicine were made

Later he was called to the home of his boyhood, to become Professor of Medicine at Leland Stanford University in California Here he continued this valuable work as a physician, teacher and investigator

Dr Hewlett possessed not only an unusual intellectual equipment and ability as an investigator, teacher and physician, but also a most attractive personality Quiet and thoughtful, and giving the impression of much reserve power and force, yet he was a most interesting and agreeable companion All the members of the early group comprising this society were his personal friends He was always interested in the younger members of this society and many of them were guided in their later careers by his writings and by his personal influence

The profession of medicine has lost in Dr Hewlett one of its ablest and most valuable members, this society has lost one of its wisest and most capable counselors

But we have lost much more, we have all lost a sincere and true friend It is therefore especially fitting for this society to record our admiration for Dr Hewlett as an able scientist, our appreciation of him as a wise and skillful physician, and at the same time express the personal affection which all of us had for him as a fellow worker and a friend

PRESIDENT'S ADDRESS

MALARIA SOME INTERESTING OBSERVATIONS RELATIVE TO MALARIA
THAT HAVE BEEN MADE IN THE TREATMENT OF GENERAL PARALYSIS
BY INOCULATION WITH MALARIA PLASMODIA

C C BASS

In 1917, Wagner von Jauregg (1) first reported the treatment of general paralysis by inoculation with malaria Favorable results were obtained in six of the nine cases, three of which were later re-

ported to be still actively at work, after four years. In 1922, he reported more than 200 cases treated by this method, over 50 of which showed remissions sufficient to permit the majority of them to resume their former occupations. His early report was followed by reports of other observers in Germany and Austria particularly, but still later in several other countries (2) (3) (4) (5) (6) (7). Reports of apparently favorable results in this disease, for which no satisfactory treatment had been available hitherto, have led to an ever increasing employment of this method until at the present time the literature contains reports of a few thousand cases. Most of the reports indicate favorable results in a proportion of cases ranging from almost none up to as high as 88 per cent (8).

On the whole, the results published are most encouraging. However, still more time and experience may be necessary to show how much allowance should be made for over enthusiasm in the use of a new and somewhat spectacular method of treatment and the difficulty of controlling the personal factor in the interpretation of the clinical state, especially in nervous and mental diseases. This applies particularly to syphilis of the nervous system in which the condition is chronic. The progress is often extremely slow, there are spontaneous improvements and the symptomatology is such that one is easily led astray. There is great danger of misinterpreting slight remissions as arrest and of attributing apparent results to treatment when they would have occurred otherwise.

Whatever therapeutic value this method of treatment may prove to have, the deliberate inoculation of hundreds and even thousands of patients with malaria under favorable conditions of control, in many particulars at least, has afforded an opportunity for observations upon certain features of malaria, far greater than had previously existed. These observations have served to emphasize certain facts in regard to malaria which were already well known but perhaps not so thoroughly appreciated and also to bring to light other facts about which much less was known. Since the observations with regard to malaria were made incidentally in most instances, when the main objective was treatment of another entirely different disease, they are probably less influenced by pre-formed ideas. My purpose is to call attention briefly to some of these points which may be called by-products of clinical investigation of the treatment of general paralysis.

Method of inoculation Blood drawn from the vein of a malaria subject has been injected intramuscularly, subcutaneously or intravenously. Inoculation directly into the blood stream is somewhat more certain to infect, although both subcutaneous and intramuscular inoculations are nearly always successful. Injection of blood immediately after withdrawal gives best results, but citrated blood has been used successfully in some instances several hours after its withdrawal. In one instance (9) defibrinated blood was kept 'on ice' for 65 hours and used successfully. By keeping it on ice, it has retained its infectivity after transportation over long distances. Inoculation by the bites of infected mosquitoes has been employed, but it has been found that on the average, cases inoculated in this way, are much more difficult to cure of their infection permanently. In one instance reported by Davidson (10) of 23 mosquito infected cases, 56.5 per cent relapsed after 30 grains quinine daily for 3 days, whereas of 33 inoculated cases only 3.3 per cent relapsed. Many others have noted the infrequency of relapse of inoculated cases of malaria. One of the facts that seems to be established by these studies is that passage of malaria parasites through the sporozoite cycle in the mosquito increases their resistance in the human body.

Incubation period The incubation period, measured by the appearance of parasites in the peripheral blood and the occurrence of fever, varies according to the mode of infection. It averages about fourteen days following infection by mosquitoes, a little longer following intramuscular or subcutaneous inoculation, and about seven days following intravenous inoculation. Clinical symptoms do not seem to be influenced by the mode of infection.

Different strains Considerable variation in different strains of the same variety of parasite with regard to the height of the temperature has been found. For instance, Bunker and Kirby (11), using two different strains of *P. vivax*, found with one that the temperature reached 105°F in 23 per cent of the cases, while with the other, 86 per cent reached this temperature. Only 3 per cent of the cases inoculated with one of the strains reached 106°F, while 64 per cent of the cases inoculated with the other one reached this height. Other effects, such as jaundice and anemia, are much more marked from certain strains than from others of the same variety of parasites.

Immunity Although it has been possible to infect nearly all cases, a few have been found that seem not to be susceptible. Bunker and Kirby (11) cite one patient who failed to acquire malaria after four widely separated intravenous inoculations although other patients inoculated with the same blood at the same time developed the disease in the usual manner. A good many cases are also reported where patients who had recovered from a successful inoculation, either spontaneously or as the result of treatment, were immune and could not be infected again. Acquired immunity therefore exists, although rarely.

Spontaneous recovery A great many cases reported have lost their infection and entirely recovered after only a few paroxysms, without any quinine or other treatment. This often occurs in the same way in naturally infected persons, as is well known to those who are familiar with malaria. In fact, in regions where malaria is very prevalent, there are a great many infected persons who have few or no symptoms, although parasites in their blood are easily demonstrated. Spontaneous recovery occurs in many of these and this is followed by more or less immunity from subsequent infection. However, we have shown (12) several years ago that whatever immunity follows spontaneous recovery is probably of short duration.

Effect of treatment The effect of treatment in these inoculated cases of malaria under favorable conditions for observation has been impressive and is probably the most important fact from a practical standpoint that has been brought out. The experiences reported emphasize the effectiveness of quinine in the treatment of malaria and indicate that the large amount often given to malaria patients is unnecessary. The treatment used by the different observers has varied considerably, ranging from 5 to 30 grains quinine (usually by mouth) daily for periods of from three days to two weeks and occasionally, but rarely, longer. Among the three or four thousand cases reported, the clinical attacks were promptly controlled, usually within 24 hours and always within two or three days. This is in line with the experience of many students of malaria to the effect that attacks of this disease are always promptly controlled by the use of moderate doses of quinine.

Clinical investigation or clinical research, the encouragement and

promotion of which is the object of this Society, has, in this instance, not only furnished information relative to what it now seems may be a valuable therapeutic agent for general paralysis, but at the same time it has shed new light on the entirely unrelated disease, malaria, and has impressively emphasized certain previously known facts relative to this disease

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The Relative Importance of the Systolic and Diastolic Blood Pressure in Maintaining the Coronary Circulation By FRED M SMITH and (by invitation) G H MILLER and V C GRABER, Iowa City

The object of the investigation was to determine the effect of specific changes in the systolic and diastolic blood pressure on the coronary circulation

Dogs were employed They were anaesthetized and the blood pressure from the carotid artery was registered on the kymograph by a Straub membrane manometer The chest was opened and a Morawitz-Zahn cannula was introduced into the coronary sinus The blood was prevented from coagulating by heparin The blood from the coronary sinus was collected in a 50 cc graduate, and the amounts were registered on the kymograph The blood was maintained at a constant temperature and reintroduced into the femoral vein Changes in the systolic and diastolic pressures were induced by constricting the thoracic aorta, and by experimentally produced arterio-venous aneurysm and aortic regurgitation

When the thoracic aorta was constricted by an adjustable clamp, it was possible, within certain limits, to increase the diastolic pressure without altering the systolic pressure The coronary flow was increased in proportion to the elevation of

diastolic pressure When the arterio-venous aneurysm was employed, the diastolic pressure was reduced and the systolic slightly elevated and the rate of coronary flow was decreased in spite of the accompanying elevation of the systolic pressure In those instances in which the diastolic pressure was reduced by puncturing the aortic valves, a similar, but even more striking reduction in coronary flow was obtained

The Relationship of the Hydrogen Ion to the Genesis of the Cardiac Rhythm By EDWARD P. CARTER and (by invitation) E. COWLES ANDRUS, Baltimore

Changes in the H ion concentration of the fluid bathing the heart is shown to affect the origin and spread of the excitatory process as follows

- 1 A change from pH 7.4 to pH 7.0 causes slowing of the rhythm, lengthens the P-R interval and depresses intra-auricular conduction

- 2 Raising the pH from 7.4 to 7.8 results in a quickening of the rhythm and an increase in the rate of conduction

- 3 Vagomimetic substances are less effective at pH 7.8 and more effective at pH 7.0 Conversely the action of sympathomimetic drugs is enhanced at pH 7.8 and depressed at pH 7.0

- 4 Studies of the results of altering the Ca concentration at constant pH indicate that the effects of pH changes cannot be due to the changes in the ionization of the Ca in solution

- 5 There is evidence that carbonic acid or its ions may exert a "specific effect" upon the rhythm. An increase in the H_2CO_3 content of the perfusate brought about without change in the pH of the solution causes an increase in the rate of spontaneous development and propagation of the excitatory process In a certain proportion of experiments, however, this effect comes on more slowly than the effects resulting from a change from a normal to a more alkaline perfusate and it tends also to pass off more gradually after return to the solution containing little or no carbonic acid The ability of carbonic acid or its ions to penetrate the cell and to raise the pH of its contents is suggested as an explanation of this "specific effect"

Upon the basis of these results the authors suggest that the rate of origin and propagation of the excitatory process in the heart is dependent upon the difference in H ion concentration within and without the cardiac cells

The Effect of Irregularity of the Rhythm of the Heart on the Blood Flow in Dogs

By H. J. STEWART, J. H. CRAWFORD and A. B. HASTINGS (by invitation) and A. E. COHN

Evidence has been obtained of the rate of blood flow in unanesthetized dogs during experimental auricular fibrillation Two insulated wire electrodes were sutured to the right auricle under aseptic technique 24 hours preceding the experiment Auricular fibrillation was induced by faradic stimulation of the auricles and rapid regular rhythms by single induced shocks at known regular rates Electrocardiograms were taken before, during and after stimulation Arterial

blood was obtained from a femoral artery. Samples of mixed venous blood were obtained by a technique devised by Stewart. Oxygen contents and capacities of the arterial and mixed venous blood were determined by means of Van Slyke's manometric method before, during and after the induced rhythms.

Summary 1 During auricular fibrillation the oxygen saturation of the arterial blood was unchanged, during regular tachycardia it was usually unchanged but was occasionally decreased 4 to 7 per cent.

2 Calculations of the relative changes in blood flow during the period of auricular fibrillation indicate decreases varying from 20 to 62 per cent of the normal.

3 The blood flow during regular tachycardia was unchanged in 11 observations in 7 dogs and decreased in 5 observations in 3 dogs.

4 In 5 experiments the same or comparable ventricular rates were obtained during auricular fibrillation and during regular tachycardia in the same dog. The blood flow was decreased in auricular fibrillation but was decreased in the regular tachycardia in only one observation.

Factors such as ventricular rate and pulse deficit concerned in determining when this difference in the circulation rate between auricular fibrillation and regular tachycardia occurs are now being analysed.

The Velocity of Blood Flow in Health and Disease By HERRMANN BLUMGART and (by invitation) SOMA WEISS, Boston

The velocity of blood flow has been determined in over one hundred and fifty patients between the ages of eighteen and seventy-five. Active deposit of radium was injected into the cubital vein of one arm. When the active deposit reached the cubital arterial vessels of the other arm, the beta particles and gamma rays emerged through the tissues of the arm and were automatically registered by an appropriate detecting device. No withdrawal of blood was necessary. No toxic effects have been noted.

In fifty normal individuals in whom the venous pressure and vital capacity were normal, the time from elbow to elbow has ranged from fifteen to twenty-four seconds. The average time was eighteen seconds and only nine individuals exceeded twenty seconds. In a given individual, successive determinations with varied dosages usually checked within two seconds, in a few instances within three seconds. Circulation times above twenty-four seconds were always associated with pathological conditions in the circulatory system. In twelve patients with auricular fibrillation the time averaged forty-four seconds with a range of twenty-eight to sixty. In four patients when sinus rhythm was induced by quinine the time was shortened, on the average, eight seconds with a range of from two to sixteen seconds.

In emphysematous patients in whom the vital capacity was conspicuously reduced, the time was normal in some and prolonged in others, according to the degree of myocardial involvement.

Observations in other diseases were made and further studies are in progress.

Studies of Vital Capacity in the Negro Race By WILSON G. SMILLIE and (by invitation) DONALD C. AUGUSTINE, New York

In the course of an investigation to determine the effect of varying intensities of hookworm infestation upon the rate of growth and development a group of about two thousand school children in south Alabama was studied. Negro children were observed as well as whites. The standard measurements for normal children developed by the U. S. P. H. Service were used, i. e., standing height, sitting height, weight, certain strength tests, vital capacity and hemoglobin.

It was discovered that normal negro children have a markedly lower vital capacity than normal white children of the same age, sex and weight. The standing height curve of growth of the two races was found to be quite similar but the trunk length or sitting height of the white children was found to be greater than that of negro children.

In order to determine whether or not the strikingly low vital capacity observed in negroes was limited to children only, a group of about two hundred strong, healthy, white and negro adults was studied. These men were all prisoners at a large convict logging camp. The results showed that adult negroes as well as negro children have a vital capacity markedly lower than whites.

Distribution of Jaundice in Circulatory Failure By JONATHAN MEAKINS, Montreal

The cases are divisible into two classes, (a) Biliary pigmentation generalized, (b) Biliary pigmentation localized to the upper part of the body, head and arms. Six cases were studied. All had severe circulatory failure and all had tricuspid insufficiency. Four cases died, with autopsy on three. One case of generalized pigmentation, which developed under observation, had acute tricuspid insufficiency with jaundice before anasarca became pronounced. The blood serum and oedema fluid of this patient were both bile stained. In five patients with localized pigmentation severe anasarca was present before serious tricuspid insufficiency and jaundice developed. Biliary pigmentation was confined to areas where oedema or ascites was not present. If hands and fore-arms were oedematous jaundice did not occur in these areas. One case extensively studied developed jaundice under observation. The blood serum was always bile stained and gave a strong direct and indirect van den Bergh reaction. The oedema fluid and the ascitic fluid was not bile stained and van den Bergh reactions on them were negative. The case studied throughout revealed a spread of the jaundice as the oedema fluid was drained away by repeated skin punctures. The questions arise whether the oedema fluid actually is not in diffusion equilibrium with the blood plasma in cardiac oedema or whether only colloids with large molecules do not diffuse. Crystalloids, urea, uric acid, creatinine and sugar are in fair equilibrium. Proteins, haemoglobin and bile pigments are much diminished or absent. Bile salts may be in equilibrium as the surface tension is practically the same in both fluids. The histopathology of the liver is distinctive and important.

Studies in Obstructive Jaundice By C H GREENE and (by invitation) M ALDRICH and A M SNELL, Rochester, Minn

Obstructive jaundice was produced experimentally in dogs by ligation of the common bile duct. Cholecystectomy was also done in some animals. The initial response after obstruction depends on the presence or absence of the gall bladder. Cholecystectomized animals show a rapid increase in biliary constituents in the blood. Jaundice is marked within twenty-four hours. When the gall bladder is intact the increase in biliary constituents in the blood is delayed and jaundice may not develop for forty-eight to seventy-two hours. This effect can be shown with bilirubin, bile salts, and dyes (phenoltetrachlorophthalein and brom-sulphalein). A maximal degree of bile retention was observed at the end of the first week, thereafter there was a gradual readjustment towards normal. The serum bilirubin tended to decrease to a level of 2 to 3 mg even though obstruction was maintained. The bile salts varied but also tended towards normal. Dye retention persisted in these dogs, though its degree sometimes became less.

There was a rapid return towards normal with the relief of the obstruction when the latter was of short duration. After long continued obstruction recovery was greatly delayed. Slight bilirubinemia, with marked increase in bile salts and dye retention were observed in one animal for several months after cholecystgastrostomy. Ascites and the late clinical picture of obstructive biliary cirrhosis were observed in one dog with complete biliary obstruction for 112 days.

Alterations in Liver Function as an Index of Toxemia in Pneumococcus Lobar Pneumonia By BENEDICT R HARRIS (by invitation of Francis G Blake), New Haven, Conn

In order to determine the effect of pneumococcus lobar pneumonia on hepatic function, dye elimination tests were done on twenty-two cases, of which 9 were due to pneumococcus Type I, 3 to pneumococcus Type II, 5 to pneumococcus Type III, and 5 to pneumococcus Type IV. Impairment of liver function was found to occur consistently, the degree increasing as the disease progressed. No definite relationship between the severity of the infection (as measured by outcome), and the degree of liver impairment was found. Following recovery liver function returned rapidly to normal, indicating no permanent injury.

Impairment of function in the Type III pneumonias with 80 per cent mortality was conspicuously less than in the Types I, II, and IV, with 23 per cent mortality.

An Isolated Case of Epidemic Influenzal Pneumonia By CHANNING FROTHINGHAM, Boston

The terms, influenza and influenzal pneumonia, are quite loosely used by the medical profession and are applied to a variety of diseases. There is no way at the present time to separate febrile upsets of unknown cause which involve the upper respiratory tract and the epidemic disease which swept through the country in 1918-19, unless the epidemic disease reaches the lungs. It is possible by a study of the lung tissue at autopsy to pick out definite lesions which are peculiar to

this epidemic disease, even when complicating organisms add to the pulmonary lesion

Isolated cases of the epidemic disease that have been proven at autopsy are so rare that it seems worth while to report these cases with the hope that as time goes on it will be possible to establish their identity during life even without the complication of pneumonia. Apparently if the epidemic disease invades the lung it is possible to make a diagnosis by the peculiarity of the x-ray findings. This case was suspected by the x-ray examination during life and confirmed by the examination of the lungs at autopsy to be a case of the epidemic disease with pulmonary complications

Changes in Serum Freezing Point and in the Concentration of Serum Electrolytes during Lobar Pneumonia By F WILLIAM SUNDERMAN and J G CAMACK (by invitation) and J H. AUSTIN, Philadelphia

The changes in electrolyte and non-electrolyte concentrations in the serum in twenty-two cases of lobar pneumonia were followed through the febrile and afebrile periods by means of freezing point, conductivity, and refractometric measurements, supplemented with total base, chloride and CO_2 analyses, and with certain non-electrolyte determinations. Whenever possible, bleedings were made at intervals of two or three days on each patient before and after the crisis

During the active infection there was a decrease in the concentration of electrolytes in the serum and a proportional decrease in the freezing point depression. After the crisis the electrolytes returned to the normal range, the total electrolytes faster than the chlorides, whereas, the freezing point depression was increased above the normal limits. This increase in freezing point depression at the time of the crisis must be due to abnormally higher concentration of non-electrolytes, the precise nature of which was not determined

A Study of Red Blood Cell Permeability By J P PETERS and (by invitation) A J EISENMAN, and A M WAKEMAN, New Haven Conn

Human blood was brought into equilibrium with 40 mm CO_2 in air at 38°C . Each sample of blood was divided into three parts. The first was analysed directly. To the second and third were added equimolar amounts of a sodium and of a potassium salt. Chloride and bicarbonate were used. Cell volume and serum protein, CO content and Cl concentration in the whole blood and the serum, and serum total base, were determined

Eight experiments were done all of which showed similar changes. The total base found in the serum to which the salt had been added agreed within 2 mM with that calculated assuming no shift of base across the membrane. The variation was not consistently above or below the calculated and it was within the limits of error for the method. There was a transfer of Cl and CO_2 across the membrane. Whether equimolar concentration of Na or K salt was added, the changes in the distribution of water, Cl and CO_2 were quantitatively identical in

the analyses. Therefore, within the limits of these experiments, no evidence of any change in the distribution of base between cell and serum was found.

The Carbon Dioxide Equilibrium in Alveolar Air and Arterial Blood during Exercise

By A. V. BOCK and (by invitation) D. B. DILL, J. S. LAWRENCE and L. M. HURATHAL, Boston

The validity of samples of alveolar CO_2 obtained by the Haldane-Priestley method during exercise has long been questioned. Data are presented showing that by means of a slight modification of the method as originally described, a close agreement between the tension of CO_2 in the alveoli and that of arterial blood can be demonstrated. This fact bears an important relationship to practically all methods now employed for the determination of the total blood flow.

The Influence of Anti-rheumatic Drugs upon the Arthritis of Serum Disease

By C. L. DERICK and CHARLES H. HITCHCOCK (by invitation) and HOMER F. SWIFT, New York

As a result of bed-side observation, and from the character of the joint fluid of patients with serum disease it seemed that the "arthralgia" might be inflammatory in nature. Attempts were made, therefore, to determine whether these symptoms might be influenced by anti-rheumatic drugs. Previous observations had shown that little effect was discernible when the drugs were administered after the onset of "arthralgia," hence in these series of cases the drugs were started shortly after the time the serum was discontinued, and were given for from 10 to 14 days. In one series the patients were treated with neocinchophen, in another with aspirin. No difference in the action of these two drugs could be detected. While the urticaria was unaffected, the amount of arthritis in these patients was markedly diminished. The rate and intensity of precipitin formation against horse serum and the persistence of horse serum in the blood of these patients was studied and compared with similar observations made by Longcope and Rackemann, and MacKenzie and Leake. As a rule the antibody in these drug-treated patients was much lower in concentration, and the antigen (horse serum) persisted longer than in the patients reported by the authors above mentioned. It is suggested, therefore, that the arthritis may depend upon the development of a certain concentration of antibody by the patient, and that the depression of antibody formation, the result of the drug administered, may be the cause of the decreased amount of arthritis.

Experimental Nephritis Produced by X-ray

By JAMES P. O'HARE and (by invitation) HUGO O. ALTNOW, THOMAS D. CHRISTIAN, JR., and ABNER CALHOUN, Boston

In the first group of rabbits incision was made over the right kidney which was drawn up into the wound and exposed once to direct action of x-ray. In different rabbits of this group, x-ray exposure varied in degree. The animals were killed

at intervals, varying from one day to four and a half months. Minor changes were noted in the rabbits exposed to smaller dosage and sacrificed early. In rabbits exposed to the larger dosage and allowed to live for three months or more, there was marked sclerosis of the kidney, reducing the organ in some cases to one-half the size of the control left kidney.

A second series was treated in a similar way except that both kidneys were exposed to the x-ray, the second kidney after the animal had fully recovered from the operation on the first kidney.

The Action of Parathyroid upon Calcium and Lead in the Bones By DONALD HUNTER (by invitation) and J. C. AUB, Boston

Patients with lead poisoning were given a calcium deficient diet containing 110 mgms. of Ca daily, so that they excreted more calcium than they ingested. The total output of calcium and lead in urine and feces was estimated.

Collip's parathyroid extract was then injected twice daily. In five out of six patients increasing doses of the extract caused a gradual increase of the blood calcium to 13 or 14 mgms. per 100 cc. The excretion of both calcium and lead then rose markedly. The lead excretion rose on several occasions to over 25 mgms. a day, a level nearly twice as high as that obtained in our previous methods of treatment. No untoward symptoms were observed.

The lead which was excreted in these observations had been stored almost entirely in the bones. The excess of calcium must have come very largely from the bones. Therefore these observations indicate that parathyroid may cause a mobilization and excretion of inorganic salts from the bones. They also contribute to the evidence that lead and calcium run parallel in their reactions in the body. This method offers a very effective but somewhat unsafe treatment for the elimination of lead.

Mechanism of the Action of Iodides on the Nitrogen Metabolism By G. P. GRABFIELD and (by invitation) C. GRAY and B. FLOWER, Boston

It has been previously shown that the administration of iodides to human subjects causes an increase in the nitrogen excretion. Hesse has shown that a similar reaction occurs in dogs.

The present work was done on dogs maintained in positive nitrogen balance on a diet containing 2.8 grams of nitrogen and a total of 660 cc. of fluid. After the nitrogen excretion had become constant sodium iodide was administered subcutaneously in doses of 0.4 grams per kilo per day for three days. During the period of drug administration the total nitrogen of the urine rose on the average 23 per cent a day, returning to the previous level after the administration was stopped. The total sulphur determined by method of Fiske remained unchanged.

After thyroidectomy the same experiment was carried out but the injection of iodides in the absence of the thyroid failed to cause an increase in the nitrogen excretion.

It is concluded, therefore, that this increase in nitrogen excretion is due not to the breaking down of the body protein but to an excretion of "deposit nitrogen" containing no sulphur and that the presence of the thyroid is necessary for this action of iodides

Blood Volume in Myxedema By W O THOMPSON (by invitation of J H Means), Boston

Nine patients with myxedema averaged 22 per cent more total plasma, 28 per cent more plasma per kilo and 25 per cent more plasma per square meter of surface area when fed thyroid than when myxedematous

From observed and calculated changes in total blood volume, it is estimated that thyroid feeding in myxedema quite commonly produces increases of 25 per cent in total volume of circulating blood

The plasma decrease on omitting the thyroid and the increase on restarting it occur much more rapidly than do the corresponding changes in the volume of cells Plasma reduction is usually well marked within ten days after omitting thyroid

A parallelism exists between basal metabolic rate and plasma volume in myxedema

Once the basal metabolism has been restored to normal further increases in the dose of thyroid up to three times the maintenance dose produce only slightly greater increases in plasma volume and basal metabolism than does the maintenance dose

The plasma volume changes in myxedema are contrasted with those in cardiac edema in which plasma volume increases with increasing edema Other differences between the two are noted

Observations on Polycythemia Vera (Erythremia) during Treatment by Phenylhydrazin By G E BROWN and H Z GIFFIN, Rochester, Minn

Seven patients with polycythemia vera have been treated by phenylhydrazin hydrochloride In addition to the clinical data the observations on these cases include among other things estimations of the blood volume and viscosity, studies in blood chemistry, functional tests on the liver and the kidneys, electrocardiographic studies, estimations of heat production, and a general study of the circulatory system under stress An effort is made to arrive at some conclusions concerning the toxicity of phenylhydrazin when administered by mouth and the significance of the jaundice and of the thrombosis which have occurred during treatment

An Investigation Relative to B Welchii Infection of the Intestinal Tract as the Etiological Factor in Pernicious Anemia By ROBERT N NYE, Boston

In confirmation of the work of others a very great increase in *B welchii* spores was found in stools from cases of pernicious anemia A similar increase was found

in stools from cases with achylia gastrica without pernicious anemia. Crude estimates of vegetative forms showed at least no more in pernicious anemia stools than in those from normal individuals

Contrary to general belief, it seems probable that the reaction of the normal small intestine, at least as far as the mid portion, possibly as far as the cecum, is decidedly on the acid side of neutrality, due chiefly to the presence of normal gastric juice. A neutral or slightly alkaline reaction is necessary for active sporulation of *B. welchii*, hence spore production in at least two thirds of the normal gastro-intestinal tract is at a minimum. However, in conditions of achylia gastrica such inhibitory influences are lacking and a marked increase in spores without increase in active or vegetative forms results. Therefore, the spore increase in pernicious anemia is probably secondary to the achylia gastrica rather than indicative that chronic intestinal infection with *B. welchii* is the etiological factor in pernicious anemia.

Studies on the Relation of Monilia to Pernicious Anemia By G. O. BROWN and (by invitation) CECILE JACOBSON and O. GARCIA, St. Louis

Rabbits and guinea pigs were inoculated with monilia isolated from the stools of active cases of pernicious anemia. Intravenous, subcutaneous and intraperitoneal injections of monilia, while causing slight decreases in the red count and hemoglobin of some animals, in no instance produced a picture approximating pernicious anemia in man. After inoculation, agglutinins and complement fixing antibodies for monilia appeared in the blood.

Animals fed large numbers of monilia by mouth, over long periods of time, developed no anemia. The organisms could be recovered from the stools while the feedings were in progress, but quickly disappeared if the feedings were discontinued.

The sera of a number of cases of pernicious anemia from which monilia have been isolated showed positive complement fixation tests with antigen prepared from monilia. The reaction is specific, in that sera giving positive Wassermann and positive tuberculosis complement fixation tests give negative results with the monilia antigen. Monilia can also be found in other conditions than pernicious anemia. Sera of a number of such cases have given positive complement fixation tests with antigen prepared from monilia isolated from a case of pernicious anemia.

Studies on Gastric Anacidity By C. S. KEEFER (by invitation) and A. L. BLOOMFIELD, Baltimore

During the course of some studies on gastric secretion a method was devised by which the volume of gastric secretion and discharge could be measured quantitatively. Applying this method to cases of gastric anacidity we have been able to obtain some information regarding the volume of secretion in such cases.

We divided the cases into three groups and compared them according to the

average volume of gastric juice secreted in 10 minute periods, with the following results

- Group A Control Series
- Group B Anacidity cases without gastric disease
- Group C Anacidity cases with gastric disease

Percentage of cases with volumes of 10 to 40 cc

Group A = 89 per cent

Group B = 93 per cent

Group C = 40 per cent

Percentage of cases with volumes of less than 10 cc

Group A = 3.5 per cent

Group B = 7.0 per cent

Group C = 60.0 per cent

It is apparent from these results that while the cases of anacidity without gastric disease have practically the same volume of secretion as the normal controls, the cases of anacidity with gastric disease have a very much smaller volume of secretion. In other words in any case of gastric anacidity in which the volume secretion is below 10 cc (average of several 10 minute periods) one is probably dealing with organic gastric disease, whereas, if volume of secretion is over 10 cc it is more likely to be a functional anacidity.

Preferential Utilization of Carbohydrates in Diabetes By WALTER R. CAMPBELL
and (by invitation) J. MARKOWITZ, Toronto

In the milder cases of diabetes there has appeared to be some grounds for the belief that certain carbohydrates are more easily utilized by the diabetic organism than other carbohydrates. Information on this point would be of great value in treatment but the possibilities of stimulation of insulin production by the carbohydrate or by exercise, of storage of the carbohydrate as fat, as blood and tissue sugar, or as glycogen, and of non-absorption from the alimentary tract, have seldom been given sufficient attention in arriving at a conclusion. By a new type of experiment an attempt has been made to throw some light on this problem. On a weighed diet, consisting of meat, raw pancreas and glucose, totally depancreatized dogs receiving insulin twice daily with meals and kept in nitrogenous equilibrium excrete a fairly constant amount of sugar daily. After a control period the carbohydrate to be tested is substituted for the glucose in the diet. After the test period a further control period using glucose is carried out. The urine is carefully collected and analysed for sugar, nitrogen and phosphates. While such a method has certain drawbacks it also has the advantages of adequate control of food intake, insulin dosage and other conditions of the experiment which are not so satisfactorily carried out on diabetic patients. The substances chosen for test were levulose, inulin, glycerin, and a triose-dihydroxyacetone. If a limited quantity of insulin permits a greater amount of the sugar to be utilized by the

animal the urinary excretion of glucose will decrease by this amount. Within the limits of error of such a method no preferential utilization of these substances was demonstrated

Diabetic Children's Diets in Different Clinics, Calculated from 532 Reports By H. GRAY, Santa Barbara

Stimulated by the tabulation by Holt and Fales of normal children's diets in grams per kilogram of body weight, a similar tabulation was undertaken for diabetic children. The great courtesy of a number of prominent students of diabetes in furnishing details on their patients, with a smaller amount from the literature or similar data at the time of a patient's discharge, has made possible this survey. For each investigator averages have been made, and the differences between these and the grand average have been examined by biometric technic to determine which differences are statistically significant. It is believed that this method reveals more precisely than is now known the diet distribution most favored in the years since insulin became available, and likewise clarifies such trends as diverge sufficiently from the average to demand discussion.

Metabolism during Fasting in the Human Subject By WILLIAM G. LENNON, Boston

Daily measurements of oxygen consumption, of nitrogen excretion, and of bicarbonate, sugar and non-protein nitrogen in the blood plasma were made during five fasting periods lasting from 6 to 15 days. Before the fasts, subjects were on a much lower protein intake than the subjects of most of the fasts reported in the literature. The amount of oxygen consumed, per kilogram of body weight, was increased throughout fasting in each case. Such increase, except under abnormal conditions of fluid intake, ran roughly parallel to nitrogen excretion.

Exaggerated Response of Sensitized Smooth Muscle to Mixtures of Sensitized Tissue and Homologous Antigen By HARRY L. ALEXANDER, and (by invitation) WM. G. BECKE and J. H. HOLMES, St. Louis

Guinea pigs were sensitized by intraperitoneal injections of antigenic substances of varying nitrogen content. Both uterine horns were suspended each in a separate Dale apparatus. In one Dale bath, a measured amount of homologous antigen was placed, and the amplitude of muscle contraction recorded. To the other, the supernatant fluid of a mixture containing the same measured amount of antigen ground with the perfused lungs of the same animal was added. The resulting muscle contraction was frequently greater than when antigen unmixed with tissue was used.

A similar exaggerated muscle response was occasionally obtained with mixtures of sensitized liver, uterine or kidney tissue and antigen. This exaggerated response was not constant. To account for this two possibilities are considered.

1. In the mixtures used, there is not always a correct proportion between the

amount of antigen and tissue This was not demonstrated because the conditions of such an experiment could not be kept constant

2 The tissue factor in the mixtures that causes exaggerated smooth muscle response may be unevenly distributed among various organs This is indicated by the discrepancies in the curves obtained by comparing the response to mixtures of lung tissue and antigen with those of other tissues and antigen under similar conditions

Observations on the Nature of "Heart-Burn" By CHESTER M JONES and (by invitation) WYMAN RICHARDSON, Boston

Twenty-nine normal subjects have been studied for the production of "heart-burn" by distension of the oesophagus at different levels with a balloon, the location of which was determined by fluoroscope Nineteen subjects noticed a hot burning sensation during distension of the lower third of the oesophagus The remainder complained of fullness or pressure Distension of the middle or upper third of the oesophagus caused a sensation of pressure in twenty-five cases and burning in only four Introduction, through a tube, of small quantities of acid, alkali, cold water, gastric contents, or barium suspension into the lower third of the oesophagus caused "heart-burn" in two-thirds of those in whom it was attempted Fluoroscopic examination of the barium column showed the presence of spasm at the level at which the liquid had been introduced, with reverse peristalsis above this level The spasm and reverse peristalsis persisted as long as the "heart-burn" lasted

In the above observations simple local distension seemed to be the cause of the sensations It is probable that "heart-burn" is analagous to referred pain and hunger contractions, and is a reflex disturbance of lower oesophageal tone, usually with abnormal peristalsis, occasioned either by local pathology or by functional or organic pathology elsewhere in the body

Oxygen Poisoning By C A L BINGER and (by invitation) J M FAULKNER and R L MOORE, New York

The present day, rather widespread, therapeutic use of oxygen makes it important for us to have more exact knowledge about its deleterious effects The history of this subject dates back to the work of Paul Bert, who showed that high atmospheric pressures of oxygen produced convulsions in animals Bert did not observe the characteristic pulmonary changes because his animals died before these changes occurred Lorrain Smith was the first to describe the lesions in the lungs His work has since been confirmed by others, notably by Karsner, who made a thorough study of the morbid anatomy of rabbits exposed to high partial pressures of oxygen

It was our purpose to inquire into the nature of the pulmonary lesions, to study the limits of oxygen concentration at which they developed and to make certain that there was no secondary bacterial invasion We were interested, too, in ob-

serving the effects of high concentrations of oxygen on other species, and in learning, if possible, the cause of death

We have found that mice, guinea pigs, rabbits and dogs all succumb to the effects of oxygen in concentrations of 80 per cent of one atmosphere or over in about 5 days. These animals at first show no untoward effects. They gradually, however, stop eating, lose weight, become dyspneic and cyanotic and then die. When they are in the advanced stage of dyspnea, removal from the high concentrations of oxygen brings on immediate death. Examination of their blood shows a marked anoxemia.

Apparently the pulmonary lesion is such as to interfere with the diffusion of oxygen and the same high concentrations which eventually kill the animals temporarily keep them alive. The characteristic pulmonary lesion is capillary dilatation and an hemorrhagic edema.

We are at present investigating the effect of similar concentrations of oxygen on amphibia and reptiles. Apparently the winter frog is able to survive high concentrations, though whether there is any statistical difference in his viability in oxygen as compared with air, we are not yet certain. We are at present engaged in the study of the effect of oxygen on the capillaries.

The Nephrotoxic Action of Ingested Cystine By A. C. CURTIS (by invitation) and L. H. NEWBURGH, Ann Arbor, Mich.

It has been shown by Newburgh and Marsh that the intravenous injection of cystine into dogs causes marked injury to the kidney. H. B. Lewis has shown that the kidneys of rabbits are injured by the introduction into the stomach of single large doses of cystine.

The former experiments are open to the criticism that the cystine entered the organism by an abnormal path. In the latter experiments, the introduction of a single large dose into the stomach of a fasting animal permits much more rapid absorption than would occur if an excess of cystine were taken in the usual way.

Our experiments deal with the effect of cystine when ingested as a constituent of the diet by the rat, and show that

1. Large doses cause a hemorrhagic nephropathy and death within a few days.
2. Doses only several times the minimal requirement cause moderate renal injury in the course of months.
3. The addition of cystine to diets in themselves too low in cystine to permit growth will, in small doses cause growth, in moderate doses inhibit growth, and in large doses produce actual loss of weight.

The Respiratory and Carbohydrate Metabolism of Dihydroxyacetone in Normal and Diabetic Individuals By EDWARD H. MASON, Montreal

Normals Comparable respiratory and blood sugar time curves after the ingestion of like doses, 25 or 50 grams, of glucose or dihydroxyacetone, in ten experiments on five cases have shown the following results:

1 The average maximum increment increase of the non-protein respiratory quotient after glucose was 0.026, while after dihydroxyacetone it was 0.271

2 The average maximum increase of heat production after glucose was 10.6 per cent as against 16.1 per cent for dihydroxyacetone

3 The blood sugar showed a lesser increment increase after the dihydroxyacetone

Diabetes Fifteen similar experiments on eight cases gave the following results

1 The average maximum increment increase of the non-protein respiratory quotient after glucose was 0.048, while after dihydroxyacetone it was 0.138

2 The average maximum increase of heat production after glucose was 11.2 per cent, while after dihydroxyacetone it was 19.4 per cent

3 The blood sugar showed a lesser increment increase after the dihydroxyacetone

Studies in Albuminuria By HILDING BERGLUND and (by invitation) WALTER SCRIVFR, Minneapolis, Minn

Factors influencing the albumin output through diseased kidneys have been studied in cases of acute and chronic nephrosis as well as in animal experiments. Both serum-albumin and serum-globulin usually escape into the urine. Albuminuria without globulinuria may occur but is evidently rare. A high albumin to globulin ratio in the urine signifies a lesser degree of abnormal permeability than a low ratio. The urinary albumin to globulin ratio is always higher than 1. The urinary ratio is independent of the corresponding ratio of the plasma.

On a high protein diet the total protein output through the kidneys may become increased as much as three times above the output during a previous or following period of low protein diet. Increased elimination of urea after urea feeding has no similar effect on the amount of urinary protein.

This phenomenon does not indicate increased kidney damage. In fact, it indicates a greater amount of protein circulating through the kidneys. This statement is based upon observations on the concentration of the plasma proteins. The protein synthesis after high protein feeding is a rapid process. In three days the plasma proteins may increase 30 per cent.

A Study of Supravital and Fixed Stained Preparations of Cells Obtained by Puncturing Diseased Lymphnodes in Man By CLAUDE E. FORKNER (by invitation) and GEORGE R. MINOT, Boston

Biopsy for diagnostic and prognostic information has certain disadvantages. Puncture of lymphnodes causes less discomfort to the patient and useful knowledge can be obtained rapidly by studying in supravital and fixed stained preparations cells so obtained. Material can be withdrawn on the barbed edges of a dental broach passing through the lumen of a needle, which has perforated the capsule of a lymphnode. The supravital technic offers a new method of approach to the study of the cell content of diseased lymphnodes. The conditions studied include

various types of lymphoblastoma, metastatic carcinoma, tuberculosis and simple hyperplasia. Carcinoma is distinguished easily. The significance of the cell pictures obtained in lymphoblastoma can be evaluated after collecting much data.

The Experimental Production of Lung Abscess By S. A. SCHLUETER, and I. F. WEIDLEIN (by invitation) and E. C. CUTLER, Cleveland, Ohio

A successful method for the experimental production of lung abscess has been devised in this Laboratory. The procedure consists in liberating into the jugular vein an artificial, infected embolus which is carried in the venous blood stream to the right heart and thence by way of the pulmonary artery to the lung.

The infected embolus is prepared as follows. A small segment of femoral vein, 6 to 8 mm. in length, is excised. One end is tied with silk. The other end is held open by three silk sutures, and various bacterial emulsions are transferred into the lumen with a platinum loop, together with a bit of paraffinized lead. A few drops of blood are then purposely added to serve as a temporary culture medium, and the material is encased with a second silk ligature about the open end. The embolus is then placed in a glass cannula, and the latter connected to a syringe filled with saline. An opening is made into the jugular vein, the cannula inserted and the embolus forced into the circulation. The lead filing serves to localize roentgenographically the final resting place of the embolus.

Success depends only on the type of organisms employed. With the proper bacteria the lesion has resulted in 100 per cent of the experiments.

Guanidine and Hypertension: Clinical and Laboratory Studies By C. P. HOWARD and (by invitation) I. M. RABINOWITCH, Montreal

The determination of guanidine compounds in the urine is with our present available methods not strictly quantitative. At best one can recover about 90 per cent of added guanidine compounds from watery solution or urine and this value is only obtained by repeated fractional crystallization which is a rather tedious procedure not lending itself to work on a clinical scale. For the present purpose, however, the method was sufficiently quantitative.

In view of the recent observations by Major and his co-workers which suggested a relationship between the diminished urinary excretion of guanidine compounds and hypertension, a series of observations were made on normal individuals and on patients suffering from hypertension.

In normal individuals marked fluctuations were found in the daily amounts of guanidine excreted. Two possible explanations of this are suggested. Like most other nitrogenous substances excreted in the urine, the daily amounts may vary. In view, however, of the constancy of creatinine excretion and the chemical relationship between this compound and methyl guanidine, it is quite possible that the total amount of guanidine bases excreted daily is constant for the individual, but that the relative proportions of the different compounds vary and that not all of these are recovered quantitatively by the method employed.

Our results on the whole are somewhat similar to those of Major. In one case in which daily studies were made, the decrease in blood pressure during the early period of the patient's rest in bed was *not* accompanied by an increased excretion of guanidine. Later when polyuria was induced and the blood pressure fell still further, excretion of guanidine compounds increased. This suggested a relationship between polyuria and the excretion of guanidine compounds.

An interesting observation was that in the series of cases with hypertension, the majority of those patients who had high diastolic blood pressure also showed diminished excretion of guanidine, whereas those with high systolic but normal diastolic blood pressure had normal excretion of guanidine.

In view of the above findings we are warned that much more must be known of the relationship between guanidine metabolism and hypertension. The results are, however, of sufficient interest to stimulate further work along these lines.

Effects of Roentgen Ray Therapy in Rheumatic Heart Disease By ROBERT L. LEVY and (by invitation) ROSS GOLDEN, New York

Cardiac involvement in rheumatic fever is frequent and of serious import. No method for its prevention is known, nor has it been possible to modify the progress of lesions initiated in the heart by the rheumatic process.

Radiotherapy is known to influence favorably certain forms of infection, it has also been employed to affect the physical character of scar tissue. These two types of lesion—exudative and proliferative—exist in the heart as the result of rheumatism.

Seven cases of rheumatic heart disease have received courses of roentgen ray therapy, two of them having been followed for one year. The dosage was calculated to yield about 10 per cent of the theoretical erythema dose in the region of the mitral valve.

Clinical improvement has been associated with striking changes in the form of the electrocardiogram in four cases. The remaining cases have thus far received insufficient treatment to warrant a report. The modification of the form of the electrocardiogram apparently associated with roentgen ray therapy suggests that an influence, presumably favorable, has been brought to bear upon the lesions in the heart muscle.

A limited number of roentgen ray treatments has been given to two patients with streptococcus viridans endocarditis without evident effect on clinical course or electrocardiogram.

The Prognostic Value of the Diazo Reaction in Chronic Nephritis By REGINALD FITZ and (by invitation) HARRY BLOTNER, Boston

Further Observations on Patients with Hypertension and Increased Basal Metabolic Rate By ERNST P. BOAS and (by invitation) S. SHAPIRO, New York

Patients with organic heart disease do not show increased heat production even

when their cardiac reserve is poor, if the determination is made under actual basal conditions. Only when they exhibit dyspnea at rest do they show increased heat production, and this is readily explained by the muscular exertion and mental distress accompanying the difficult respiration. We examined twenty-eight patients with essential hypertension and two with chronic glomerulonephritis and hypertension under basal conditions. None of them was dyspneic at rest. The patients with glomerulonephritis had normal heat production. Ten patients with essential hypertension exhibited basal rates above plus 15. Two or nine males and eight of nineteen females showed increased heat production. The distinguishing clinical features of these patients with high basal rates are hypertension, tachycardia, loss in weight, pigmentation of the skin and nervousness. They do not present the picture of Graves' syndrome. Partial thyroidectomy does not seem to improve their clinical condition, and iodine administration does not alter their metabolism. High diastolic pressures are not seen in Graves' disease. We believe that our observations warrant the conclusion that these patients present a special syndrome which should be distinguished from the ordinary form of hypertension as well as from Graves' Disease.

The Significance of Electrocardiograms of Low Voltage By HOWARD B. SPRAGUE (by invitation) and PAUL D. WHITE, Boston

The amplitude of the QRS waves of the human electrocardiogram is probably roughly indicative of myocardial power in cases in which the excursion is less than 5 mm. from the base line. A series of clinical cases is presented in which the electrocardiograms are correlated with the etiology of the heart disease, with the physical findings, and with the prognosis. The most important groups are the cases of arteriosclerosis, and those with myxedema or cretinism. Diminution in the intensity and change in the quality of heart sounds are frequent findings. Aberration of the electrical complexes with changes in the T wave are very common.

Electrical waves of low voltage, except in myxedema and cretinism are usually a bad prognostic sign. They are often found in serious angina and may accompany coronary occlusion. Only 10 per cent of the total cases in our series are able to be actively at work.

Intermittent Incomplete Bundle Branch Block By F. A. WILLIAMS (by invitation) and Norman M. Keith, Rochester, Minn.

Three cases are reported, each differing somewhat in type and degree, in which electrocardiograms have been obtained conforming to those of incomplete bundle branch block. In all instances the graphic changes have been present intermittently. These cases are fully discussed with reference to the electrocardiographic abnormalities. The literature on this subject is also considered.

Experimental Study of the Synovial Fluid of Patients with Arthritis and Syphilis

By ALAN M. CRESNEY and (by invitation) JAROLD E. KEMP, Baltimore

Joint fluid was aspirated from a series of ten patients in various stages of

syphilis, in whom arthritis was present in addition. The fluids were studied with reference to (a) cytology, (b) Wassermann reaction, (c) presence of bacteria by smear, culture and guinea pig inoculation, (d) presence of *Treponema pallidum* by dark field examination and intratesticular inoculation in rabbits.

Virulent strains of *T. pallidum* were obtained by rabbit inoculation from three of the ten joint fluids studied. A relatively high lymphocytosis was encountered in two of these fluids as well as in two others in which there was reasonably good evidence that the arthritis was syphilitic in origin. Fluids from those cases which were believed to be non-syphilitic did not show this relative increase in lymphocytes.

The Physiology of the Leucocytes in the Saliva in Health and Disease By RAPHAEL ISAACS, Boston

Quantitative studies of the leucocytes of the blood and of the saliva show that when the leucocytic blood count rises, the number of leucocytes in the saliva tends to decrease, and when the leucocytic blood count decreases, the saliva count increases. Normally, the saliva collected from the salivary ducts has few or no leucocytes. When the saliva is allowed to collect in a pool in the mouth it washes together leucocytes which have wandered through the mucous membrane. The number varies from five or less per cubic millimeter to about 150, but may reach 400 in health. When a "digestive leucocytosis" fails to show in the blood, it is frequently replaced by an increase in the number of leucocytes in the saliva. All types of leucocytes appear in the saliva, and they live for varying lengths of time dependent on the disease or the therapy. In chronic myelogenous leukemia, the saliva count may be surprisingly low when the blood count is high. After roentgen ray treatment the saliva cell count increases, at times to 10,000 or more per cubic millimeter, with the decrease in the blood leucocyte count. In aleukemic leukemia the saliva cell count may be very high.

The Treatment of Paroxysmal Auricular Tachycardia By S. A. LEVINE and (by invitation) HARRY BLOTNER, Boston

The Onset of Digitalis Effect after Intravenous Administration By HAROLD E. B. PARDEE, New York

The effects of digitalis which were observed in these experiments were (1) the slowing of ventricular rate in patients with rapid rates from auricular fibrillation, (2) the diminution in size of the T wave of patients with normal rhythm.

Digitalan solution and Digalen were the chief preparations used. These were given intravenously in a dose of 1 mm. of the solution per pound of body weight of the patient. In the patients with auricular fibrillation the heart rate was counted with a stethoscope several times for one-half minute periods to obtain a control rate and then at various intervals during the next 24 hours. In those with normal rhythm a control electrocardiogram was taken before the drug was given, and then at proper intervals for 24 hours.

It was found that the effect upon rate often began as soon as 5 minutes after intravenous administration of these doses, always within 15 minutes. The slowing progressed rapidly during the first 30 minutes and somewhat less rapidly during the next hour. The maximum slowing was usually 2 hours after administration but most of the effect developed during the first hour. After reaching its maximum the effect persisted, diminishing slightly, but even 24 hours after the dose, the rate had not reached its control level.

The effect upon the T wave also appeared as soon as 5 minutes after the dose and reached its maximum about 2 or 3 hours later, then passed off gradually.

The important results of these experiments are to show that the doses of these preparations previously suggested by the author are proper for clinical use, that the onset of digitalis effects after proper intravenous doses is more prompt than was previously believed and that the effect upon rate and the effect upon the muscle proceed at about an equal rate.

Rickets in Dogs By ALFRED T. SHOHL, New Haven

Observations on the pathogenicity of Brucella abortus for human beings By ERNEST C. DICKSON, San Francisco

Following an illness in January, 1925, Miss C. was advised to drink plenty of milk. From February 15 to June 7 she consumed from four to six glasses of milk daily, part of it being pasteurized and the remainder being raw milk from an inspected dairy.

In the latter part of May she developed an intermittent fever associated with sweating, fatigue and pains in the legs. The fever persisted for several weeks ranging from 99 to 101 or 102 degrees daily with occasional remissions. The only notable physical sign was a palpable spleen.

On July 3, the patient's blood serum caused complete agglutination of *Br. melitensis* in dilution of 1:500 and partial agglutination in 1:1000. Subsequently Miss Alice Evans at Washington reported that it was the *Br. abortus* variety.

Investigation of the dairy showed that there had been little trouble with infectious abortion although two cows had aborted within fifteen months. Whey prepared from the milk of these cows caused agglutination of *Br. abortus* in dilutions of 1:100 and 1:200, respectively, and guinea pigs which were injected with the milk developed typical lesions of *Br. abortus* infection from which *Br. abortus* was recovered in pure culture.

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